The Great Lakes Entomologist

Volume 53
Numbers 1 & 2 - Spring/Summer 2020 *Numbers 1 & 2 - Spring/Summer 2020*

Article 1

# Full issue for TGLE Vol. 53 Nos. 1 & 2

Vol. 53, Nos. 1 & 2 Spring/Summer 2020

# THE GREAT LAKES ENTOMOLOGIST

*PUBLISHED BY*

THE MICHIGAN ENTOMOLOGICAL SOCIETY

## THE MICHIGAN ENTOMOLOGICAL SOCIETY 2019–20

### OFFICERS

| | |
|---|---|
| President | Elly Maxwell |
| President Elect | Duke Elsner |
| Immediate Pate President | David Houghton |
| Secretary | Adrienne O'Brien |
| Treasurer | Angie Pytel |
| Member-at-Large | Thomas E. Moore |
| Member-at-Large | Martin Andree |
| Member-at-Large | James Dunn |
| Member-at-Large | Ralph Gorton |
| Lead Journal Scientific Editor | Kristi Bugajski |
| Lead Journal Production Editor | Alicia Bray |
| Associate Journal Editor | Anthony Cognato |
| Associate Journal Editor | Julie Craves |
| Associate Journal Editor | David Houghton |
| Associate Journal Editor | Ronald Priest |
| Associate Journal Editor | William Ruesink |
| Associate Journal Editor | William Scharf |
| Associate Journal Editor | Daniel Swanson |
| Newsletter Editor | Crystal Daileay and Duke Elsner |
| Webmaster | Mark O'Brien |

The Michigan Entomological Society traces its origins to the old Detroit Entomological Society and was organized on 4 November 1954 to ". . .promote the science of entomology in all its branches and by all feasible means, and to advance cooperation and good fellowship among persons interested in entomology." The Society attempts to facilitate the exchange of ideas and information in both amateur and professional circles, and encourages the study of insects by youth. Membership in the Society, which serves the North Central States and adjacent Canada, is open to all persons interested in entomology.

There are five paying classes of membership:
Active—annual dues $25.00
Student (to 12th grade)—annual dues $12.00
Institutional—annual dues $45.00
Sustaining—annual contribution $35.00 or more
Life—$500.00

Dues are paid on a calendar year basis (Jan. 1–Dec. 31). Memberships accepted before July 1 shall begin on the preceding January 1; memberships accepted at a later date shall begin the following January 1 unless the earlier date is requested and the required dues are paid. All members in good standing receive the Newsletter of the Society. All active and sustaining members may vote in Society affairs. All dues and contributions to the Society are deductible for Federal income tax purposes.

### SUBSCRIPTION INFORMATION

The journal is published online and open access, there is no subscription needed. Articles are available in pdf format and can be printed from the website, free of charge. To view current and past volumes, please visit http://scholar.valpo.edu/tgle/ .

# Protection of Ash Trees Under Extended Emerald Ash Borer Pressure

Fredric Miller[1,*] and Dan Mueller[1]

[1] The Morton Arboretum, 4100 IL-53, Lisle, IL 60532

[*] Corresponding author: (e-mail: fmiller@jjc.edu)

**Abstract**

Ten studies were conducted in northeastern Illinois from 2007 to 2015 to evaluate treatment formulations, rates, and application timing and methods for protection of green (*Fraxinus pennsylvanica*), white (*F. americana*) and blue ash (*F. quadrangulata*) trees from the emerald ash borer (EAB), *Agrilus planipennis* Fairmaire (Coleoptera: Buprestidae). Annual mid-May, June, July, and September basal soil drenches, basal broadcast applications, and basal trunk spray applications of imidacloprid, clothianidin, dinotefuran used alone, imidacloprid + clothianidin, dinotefuran + clothianidin, and trunk injections of emamectin benzoate were evaluated. Imidacloprid applied alone at 0.57 g a.i./2.54 cm dbh or greater, clothianidin and dinotefuran alone at 0.93 g a.i./2.54 cm dbh or greater, imidacloprid + clothianidin at 0.57 g a.i. + 0.28 g a.i.2.54 cm dbh or greater, dinotefuran + clothianidin at 0.47 g a.i. + 0.46 g a.i./2.54 cm dbh or greater, or emamectin benzoate applied at 0.2 to 0.6 g a.i./2.54 cm dbh provided good protection of ash trees up to 61 cm mean dbh. Canopy thinning was strongly correlated with the number of larval galleries/m$^2$ ($R^2 = 0.95$; $P < 0.001$) and adult EAB exit holes per m$^2$ of branch surface area ($R^2 = 0.94$; $P = 0.002$). Severe drought conditions may have contributed to a differential PCL response for treated large green ash trees growing in narrow residential parkways compared to trees growing in open park-like-landscape settings. Choice of active ingredient(s), product formulation(s), application methods and timing, EAB pressure, host susceptibility, and abiotic factors, and their role in implementing an EAB pest management plan are discussed.

**Keywords:** ash, clothianidin, dinotefuran, emamectin benzoate, emerald ash borer, imidacloprid

---

The emerald ash borer (EAB), *Agrilus planipennis* Fairmaire (Coleoptera: Buprestidae) was initially identified in June, 2002 from beetles collected in the Detroit, Michigan area (Haack et al. 2002, Cappaert et al. 2005, Herms and McCullough 2014). Green ash (*Fraxinus pennsylvanica*) and white ash (*F. americana*) are commonly planted as landscape and parkway trees throughout the eastern and midwestern United States comprising 10% to greater than 30% of the urban forest tree canopy (Raupp et al. 2006). More recently, blue ash (*F. quadrangulata*) is being considered for parkway and landscape plantings (Dirr 2009; author's personal communication with green industry professionals). Currently, chemical treatments are the only effective method for protecting existing ash trees from this insect (Poland and McCullough 2006). Costs associated with ash tree preservation and protection or removal falls on municipalities and property owners (Sydnor et al. 2007, Kovacs et al. 2010, McCullough and Mercader 2012, Creticos 2013). Early on, insecticide trials for EAB were not totally successful, consistent, or reliable, and led to widespread skepticism and rejection by arborists, urban foresters, government decision makers, and property owners as an effective EAB management strategy (Herms et al. 2009). More recently, neonicotinoid insecticide field studies have demonstrated more consistent and reliable means for protecting ash trees from EAB (Cappaert et al. 2005; Smitley et al. 2008, 2010a, b, 2015; Herms et al. 2014; Bick et al. 2018; McCullough et al. 2019; Robinette and McCullough 2019). Recent studies have demonstrated that the cost of tree removal commonly exceeds the cost of insecticide treatment (Sydnor et al. 2007, Kovacs et al. 2010, Sadof et al. 2011, McCullough and Mercader 2012, McKenney et al. 2012, Vannatta et al. 2012, Hauer and Peterson 2017). In addition, trunk injections of emamectin benzoate, used every two to three years, have been shown to be highly effective in protecting ash trees from the emerald ash borer (Smitley et al. 2010a; McCullough et al. 2011, 2019; McCullough and Mercader 2012; Herms et al. 2014; Flower et al. 2015; Lewis and Turcotte 2015; Bick et al. 2018) and are being used by green industry professionals and as general use products for use

by homeowners. Other treatment application methods including soil injections, basal soil drenches, basal broadcast applications, and basal trunk sprays of neonicotinoid class insecticides are alternatives available for use by professionals and homeowners alike for EAB management. These treatment methods require minimal equipment, are easier to apply compared to soil and trunk injection methods, and have been shown to be efficacious for both homeowners and green industry professionals for protecting ash trees up to 38 cm dbh (Smitley et al. 2010a, b; McCullough et al. 2011, 2019; Herms et al. 2014, 2019; Bick et al. 2018).

For larger trees over 38 cm dbh, the use of soil injections and basal soil drenches of neonicotinoid insecticides have not always proven to be effective for trees over 38 cm dbh and when under intense EAB pressure (Smitley et al. 2010b). Early on, rates and practices of applying imidacloprid specified a linear relationship between tree dbh and application rates. However, research by LeGoff and Ottorini (1996) and McCullough and Siegert (2007) have shown that as ash tree dbh doubles, tree surface area increases five-fold. These findings suggest the need to increase treatment rates for larger trees to account for the larger surface area and phloem biomass (Smitley et al. 2010b). With EPA approval of the 2× rate of imidacloprid for ash trees with trunk diameters greater than 38 cm, and the availability of additional neonicotinoid insecticides and emamectin benzoate, more reliable protection of ash trees over 38 cm dbh is a possibility. More recent studies by Smitley et al. (2015) and Bick et al. (2018) have shown that a spring and/or fall application of imidacloprid at the 2× rate can be effective in protecting trees over 38 cm dbh from EAB. Therefore, the objectives of this study were to evaluate the efficacy of systemic insecticides (imidacloprid, dinotefuran, clothianidin, and emamectin benzoate) and their combinations for control of the emerald ash borer (EAB) on green, white, and blue ash trees greater than 38 cm dbh by comparing various modes of application (soil application, basal bark spray, trunk injection), rate and number of applications, and timing. More specifically, we evaluated the efficacy of 1× and 2× rates of basal soil drenches of imidacloprid applied alone or in combination with clothianidin plus a 2-1-1 fertilizer; basal broadcast applications of imidacloprid in combination with a 2-1-1 fertilizer; basal soil drenches of clothianidin and dinotefuran each applied alone or in combination; a basal broadcast application of dinotefuran; basal trunk sprays of clothianidin and dinotefuran each applied alone; and trunk injections of emamectin benzoate.

## Materials and Methods

Ten different studies, each of at least four years' duration, and consisting of three to seven different treatments per study, were conducted between 2007 and 2015 on green, white, and blue ash parkway and park trees at sites in the greater Chicago, Illinois area. Depending on tree availability, five to ten single tree replicates were established per treatment rate per study site along with an equal number of untreated controls. Trees at each study site were randomly assigned a treatment or were designated an untreated control. Only healthy, pest and disease free, and undamaged trees were selected, and all study trees were in good condition at the beginning of their respective studies. The only abiotic event was the unforeseen 2012 drought which impacted all of the trees in seven of the ten studies. Six of the ten studies included trees less than 50 cm dbh, and four studies included trees greater than 50 cm dbh. Here, trees less than 50 cm dbh less will be treated as smaller trees and trees greater than 50 cm will be considered large. All study trees were evaluated in June and August of each year for percent canopy thinning (nearest 10%) by two individual evaluators as described by Smitley et al. (2008) except for the Homewood and Fermi Lab Village study sites, which were evaluated only once per season. Percent canopy loss (PCL) is used for comparing insecticide efficacy, application methods and timing, and insecticide formulations. A stand-alone fertilizer treatment was not included in the trials because previous studies have shown fertilizer treatments have no effect on ash resistance to EAB (Tanis and McCullough 2015). All Merit and Bayer Advanced Tree and Shrub (BATS) products were formulated by Bayer Corp. (Research Triangle Park, NC, U.S.), Xytect products were formulated by Rainbow Treecare Scientific Advancements, (Minnetonka, MN, U.S.), Safari products by Valent Corporation (Walnut Creek, CA, U.S.), and the TreeAge product was formulated by ArborJet Inc. (Woburn, MA, U.S.). A complete listing, by study site, of the number of single tree replicates (N), chemical treatments by trade name or acronym, rate (total active ingredient per cm dbh) applied per year, application method, and application timing is presented in Table 1. All treatments are identified in the narrative and data tables using trade names and/or acronyms, and their corresponding percent active ingredient.

**Skokie, Illinois Study (2007–2011):** Thirty large (mean dbh = 58 cm; range = 51–89 cm) green ash parkway trees, growing in Skokie, Illinois, were used to evaluate 1×, 1.5×, and 2× rates of imidacloprid applied

**Table 1. List of chemical treatments by study site, number of single tree replicates per treatment (N), trade name or acronym, common name, rate of active ingredient applied per year, and application method and timing.**

| Study site | N | Trade name Or Acronym | Common name | Rate per year (A.I. g/2.54 cm dbh) | Application method and timing |
|---|---|---|---|---|---|
| **Skokie (SK) (2007–2011)** | | | | | |
| **Mean dbh = 58 cm (range=51–89 cm)** | | | | | |
| | 10 | Xytect 75WSP | imidacloprid | 0.56 | BSD[8] (one 1X application in mid-May) |
| | 10 | Xytect 75WSP | imidacloprid | 0.84 | BSD (one 1.5X application in mid-May) |
| | 10 | Xytect 75WSP | imidacloprid | 1.12 | BSD (one 2X application in mid-May) |
| **Aurora (AU) (2009-2012)** | | | | | |
| **Mean dbh = 43 cm (range= 30-66 cm)** | | | | | |
| | 10 | BATSC[1] | imidacloprid + clothianidin | 0.58 + 0.30 | BSD (one mid-May and one mid-June application) |
| | 10 | BATSGF1[2] | imidacloprid + clothianidin | 0.76 + 0.38 | BBA[9] (one mid-May and one mid-June application)+ 2-1-1 fertilizer |
| | 10 | BATSC2X[3] | imidacloprid + clothianidin | 1.16 + 0.58 | BSD (one 2X application in mid-May) |
| **Homewood (HW) (2008-2011)** | | | | | |
| **Mean dbh = 41 cm (range= 36-61 cm)** | | | | | |
| | 10 | Merit 2F | imidacloprid | 0.58 | BSD (one mid-May application) |
| | 10 | Merit 2F | imidacloprid | 0.58 | BSD (one mid-Sept. application) |
| | 10 | BATSC | imidacloprid + clothianidin | 0.58 + 0.30 | BSD (one mid-May application) |
| | 10 | BATSGF2[4] | imidacloprid + 2-1-1 fertilizer | 0.51 | BBA (one mid-May application) |
| | 10 | BATSGF3[5] | imidacloprid + clothianidin +2-1-1 fertilizer | 0.58 + 0.30 | BBA (one mid-May application) |
| **Riverside (RV), Hinsdale (HI), Naperville (NV), and Woodridge (WR) (2012-2015)** | | | | | |
| **Mean RV dbh=61cm (range=51–91 cm) and Mean dbh = 26 cm (range = 18.8-36 cm); Mean HI dbh=51 cm (range=46–91); Mean NV dbh=58 cm (range=51-64); Mean WR dbh=37 cm (range=25-48 cm)** | | | | | |
| | 10 | Merit 2F | imidacloprid | 1.16 | BSD (one mid-May application at 2X rate) |
| | 10 | BATSC | imidacloprid + clothianidin | 0.58 + 0.30 | BSD (one mid-May application at 2X rate) |
| | 10 | BATSGF1 | imidacloprid + clothianidin | 0.76 + 0.58 | BBA (one mid-May application at 2X rate)+2-1-1 fertilizer |
| | 10 | BATSGF4[6] | imidacloprid | 0.92 + fertilizer | BBA (one mid-May application at 2X rate) |
| | 10 | BATSCF[7] | imidacloprid | 1.09 + fertilizer | BSD (one mid-May application at 2X rate) |

**(Continued on next page)**

**Table 1. (Continued)**

| Study site | N | Trade name Or Acronym | Common name | Rate per year (A.I. g/2.54 cm dbh) | Application method and timing |
|---|---|---|---|---|---|
| **Glen Ellyn (GE) (2010-2014)** | | | | | |
| **Mean dbh=41 cm (range=28-48 cm)** | | | | | |
| | 7 | Arena 50WDG | clothianidin | 0.94 | BSD (one application in mid-June) |
| | 7 | Safari 20SG | dinotefuran | 0.94 | BSD (one application in mid-June) |
| | 7 | Safari 20SG + Arena 50WDG | dinotefuran + clothianidin | 0.48 + 0.47 | BSD (one application in mid-June) |
| | 7 | Safari 20SG | dinotefuran | 0.94 | BSD (one application in mid-July) |
| | 7 | Arena 50WDG | clothianidin | 0.94 | BTS[10] (one application in mid-June) |
| | 7 | Safari 20SG | dinotefuran | 0.94 | BTS (one application in mid-July) |
| | 7 | Safari 2G | dinotefuran | 0.94 | BBA (one application in mid-June) |
| **Fermi Lab Village (FLV) (2008-2015)** | | | | | |
| **Mean dbh=44 cm (range=38-50) for small trees; Mean dbh=60 cm (range 50-70 cm) for large trees** | | | | | |
| | 10 | TREE-age (small) | emamectin benzoate (4%) | 0.2 | T1[11] Quik-Jet (one mid-May injection) |
| | 10 | TREE-age (small) | emamectin benzoate (4%) | 0.4 | TI Tree IV (one mid-May injection) |
| | 10 | TREE-age (large) | emamectin benzoate (4%) | 0.3 | TI Quik-Jet (one mid-May injection) |
| | 10 | TREE-age (large) | emamectin benzoate (4%) | 0.6 | TI Tree IV (one mid-May injection) |

**Treatment Code**

[1]BATSC = Bayer Advanced Tree and Shrub Protect and Feed Concentrate II (one mid-May and one mid-June application)
[2]BATSGF1 = Bayer Advanced Tree and Shrub Protect and Feed Granule II + 2-1-1 fertilizer (one mid-May and one mid-June application)
[3]BATSC2X = Bayer Advanced Tree and Shrub Protect and Feed Concentrate II (one mid-May application at 2X rate)
[4]BATSGF2 = Bayer Advanced Tree and Shrub Protect and Feed Granule II + 2-1-1 fertilizer (one mid-May application)
[5]BATSGF3 = Bayer Advanced Tree and Shrub Protect and Feed Granule II + 2-1-1 fertilizer (1.1% applied as one mid-May application at 0.51 g a.i./cm dbh)
[6]BATSGF4 = Bayer Advanced Tree and Shrub Protect and Feed Granule II + 2-1-1 fertilizer (1.1% applied as one mid-May application at the 2X rate at 0.58 g a.i./cm dbh)
[7]BATSCF = Bayer Advanced Tree and Shrub Protect and Feed Concentrate II + 2-1-1 fertilizer (1.47% applied as one mid-May application at 2X rate)
[8]BSD = Basal soil drench
[9]BBA = Basal broadcast application
[10]BTS = Basal trunk spray
[11]TI = Trunk injection

as a basal soil drench. All three treatments were applied annually in mid-May as a basal soil drench containing Xytect 75WSP (imidacloprid 75%) at either 0.56 gm (1× rate), 0.84 gm (1.5× rate), or 1.12 gm (2× rate) a.i./2.54 cm dbh. Ten single tree replicates were used per treatment rate and an additional ten trees served as untreated controls. Depending on existing soil moisture conditions, the product was diluted in 8–16 L of water and poured around the base of the trunk.

**Aurora, Illinois Study (2009–2012):** Thirty green ash parkway trees, with a mean dbh of 43 cm (range = 30–66 cm), growing in Aurora, Illinois, were treated with three different treatments with ten single tree replicates per treatment rate. An additional ten trees served as untreated controls. The three treatments included a single basal soil drench application of a homeowner formulation of BATSC2X (imidacloprid 0.74% + clothianidin 0.37%) at the 2× rate in mid-May; two basal soil drench applications of a homeowner formulation of BATSC (imidacloprid 0.74% + clothianidin 0.37%) at the 1× rate made in mid-May and again in mid-June, and two basal broadcast applications of a homeowner formulation of BATSGF1 (imidacloprid 0.76% + clothianidin 0.38% plus a 2-1-1 fertilizer) made in mid-May and again in mid-June. The two BATSC basal soil drench treatments were diluted in 4–12 L of water, depending on existing soil moisture conditions, and applied to the soil around the trunk base. The BATSGF1 basal broadcast application was applied evenly on the soil surface within 1 m of the tree trunk, and watered in with 4 L of water immediately after application.

**Homewood, Illinois Study (2009–2012):** Fifty green ash parkway trees, with a mean dbh of 41 cm (range of 36–61 cm), growing in Homewood, Illinois, were treated with five different treatments with ten single tree replicates per treatment rate. Ten additional trees served as untreated controls. The five treatments included either one single mid-May or one single mid-September basal soil drench of professional Merit 2F (imidacloprid 25%); one single mid-May basal soil drench of a homeowner formulation of BATSC (imidacloprid 0.76%+clothianidin 0.58%); or one single mid-May basal broadcast application of either a homeowner formulation of BATSGF2 (imidacloprid 1.1% plus a 2-1-1 fertilizer) or BATSGF3 (imidacloprid 0.55% + clothianidin 0.275% plus a 2-1-1 fertilizer). The basal soil drenches of professional Merit 2F and BATSC were applied by diluting the product in approximately 4.0 liters of water and drenching evenly around the base of the trunk. The basal broadcast application treatments were applied evenly in a circle within 1 m of the tree trunk and immediately watered in with 4 L of water.

**Riverside, Hinsdale, Naperville, and Woodridge, Illinois Studies (2012–2015):** The Riverside site included 60 large (mean dbh = 61 cm; range = 51–91 cm) green ash trees growing in a park setting, and the Hinsdale site included 60 large parkway green ash trees (mean dbh = 51 cm; range = 46–91 cm). There were ten single tree replicates for each of the five treatment rates, and an additional ten trees served as untreated controls at each site. The Riverside blue ash study site consisted of 36 blue ash parkway trees (mean dbh = 26 cm (range = 19–36 cm) with six single tree replicates for each of the five treatment rates, and an additional six trees served as untreated controls.

The Naperville and Woodridge studies focused on the protection of 25 white ash parkway trees at each site with a mean dbh of 58 cm (range = 51–64 cm) and 37 cm (range = 25–48 cm), respectively. There were five single tree replicates for each of the five treatment rates, and an additional five trees at each site served as untreated controls. The five treatments were applied at the 2× rate at each of the five study sites (i.e. Riverside, Hinsdale, Naperville, and Woodridge), and consisted of an annual mid-May basal soil drench of professional Merit 2F (imidacloprid 25%), a mid-May basal soil drench homeowner formulation of either BATSC (imidacloprid 0.76% + clothianidin 0.58%) or BATSCF (imidacloprid 1.47% plus a 2-1-1 fertilizer), and basal broadcast applications of a homeowner formulation of either BATSGF1 (imidacloprid 0.76% + clothianidin 0.58% + 2-1-1 fertilizer) or BATSGF4 (imidacloprid 1.1% plus a 2-1-1 fertilizer). The basal soil drench applications were applied as previously described by diluting the product in approximately 4 L of water and drenching evenly around the base of the trunk. The basal broadcast applications were applied by distributing the product evenly within 1 m from the base of the tree and watering it in with 4 L of water immediately after application.

**Glen Ellyn, Illinois Study (2010–2014):** 49 green ash parkway trees, with a mean dbh of 41 cm (range = 28–48 cm), were treated with seven different treatments with seven single tree replicates per treatment or treatment combination rate. An additional seven trees served as untreated controls. Treatments included annual mid-June basal soil drenches of Arena 50WDG (clothianidin 50%) used alone, Safari 20SG (dinotefuran 20%) used alone, and Safari 20SG (dinotefuran 20%) plus Arena 50WDG (clothianidin 50%), and an annual mid-July basal soil drench treatment of Safari 20SG (dinote-

furan 20%). The basal soil drenches were mixed with water and applied within 0.5 m around the base of the trunk at a rate of 1 L of drench solution per 2.54 cm dbh. Basal trunk sprays of Arena 50WDG (clothianidin 50%) or Safari 20SG (dinotefuran 20%) were applied annually in mid-June or in mid-July, respectively to the trunk until runoff between the soil line and 1.5 m above the soil line, at a rate of approximately 20 ml/2.54 cm dbh using a Solo hand pump sprayer at 10-20 PSI. The single basal broadcast application of Safari 2G (dinotefuran 2%) was applied annually in mid-June, evenly to the soil, within 1m of the trunk and watered in with 4 L of water.

**Fermi Lab Village, Batavia, Illinois Study (2008–2015):** This eight-year study included a total of 60 green ash trees growing in residential and park areas of Fermi Lab Village (FLV) on the grounds of the Fermi National Accelerator Laboratory (FNAL) at Batavia, Illinois. The study was designed to evaluate the Quik-jet™ and Tree IV™ trunk injection systems of Tree-age (4% emamectin benzoate) for protection of green ash trees. There were ten single tree replicates for each of the four treatment rates, and an additional ten trees served as untreated controls, for each of the two dbh size classes. In mid-May 2008, 40 green ash study trees were selected for treatment and were divided into two dbh size classes (38 to 50 cm, and greater than 50 cm dbh) for a total of 20 trees in each dbh size class treatment group, and were trunk injected. The trees in the small dbh size class treatment group had a mean dbh of 39 cm (range of 36 to 46 cm) and trees in the larger dbh size class treatment group had a mean dbh of 55 cm (range of 48 to 66 cm). Within the small (38 to 50 cm dbh) size class treatment group, ten single tree replicates received a Quik-jet™ trunk injection of Tree-age (4% emamectin benzoate) at 0.2 g a.i. per 2.54 cm dbh in 5 ml of water per 2.54 cm dbh, and a second group of ten trees received a Tree IV™ trunk injection of 0.4 g a.i./2.54 cm dbh in 10 ml of water per 2.54 cm dbh. For the group of 20 larger trees (greater than over 50 cm dbh), ten single tree replicates were trunk injected with Tree-age (4% emamectin benzoate) at 0.3 g a.i. per 2.54 cm dbh in 7.5 ml of water per 2.54 cm dbh with the QUIK-jet™ trunk injection system, and a second group of ten trees received 0.6 g a.i.. per 2.54 cm dbh in 15 ml of water per 2.54 cm dbh using the Tree IV™ trunk injection method. Injection holes were drilled into the tree to a depth of approximately 5.1 cm and a plastic septum (Arborjet #4 plug) was inserted into the trunk at 20–30 cm above the ground. The number of injection sites per tree was determined by taking the dbh and dividing by two. Injection sites were spaced approximately every 15 cm around the trunk circumference. In 2007, an EAB infestation was first detected in green ash trees in the NW corner of the FNAL property. The Fermi Lab Village (FLV) study site was situated approximately 1.2 km east to southeast of the initial EAB infestation so, in mid-May, 2008, ten green ash EAB trap trees were established along a NW to SE transect to monitor for EAB spread and pressure into the FLV study site. At the end of the 2009 and 2010 field seasons, the trap trees and branches from declining portions of untreated non-study FLV trees were felled or removed, peeled, and examined for evidence of EAB galleries and life stages. All 40 of the original study trees were retreated in mid-September, 2012 using the same rates, volume of solution, number of injections sites, and application methods. The 2012 re-application treatments were delayed until mid-September, 2012 due to a record-setting regional drought which prevailed from October, 2011 through August, 2012.

**Phloem Utilization by EAB Larvae:** Assessments of phloem utilization by EAB larvae were conducted during the winter of 2011-2012 at the Aurora, Homewood, and Skokie study sites by taking branch samples from remaining untreated control trees. Two untreated trees at the Fermi Lab Village site had to be removed due to hazard and new construction and were also used for branch sampling. At all four sites, branch samples were taken at mid-canopy from each of the four cardinal directions (N, S, E, W) and branches ranged from 5 to 13 cm. in diameter and 1.2 to 1.5 m long. Samples were transported back to the Morton Arboretum entomology laboratory and peeled using a draw knife. Following peeling, measurements of EAB larval gallery area per branch, the diameter and circumference of both ends of the branch, length of the branch, the total number of galleries per branch, and the total number of adult EAB exit holes per branch were recorded. EAB gallery area in $cm^2$ was determined by measuring the length of the gallery multiplied by the mean width (width at the initiation and at the cessation of the gallery). Total available phloem surface area of each branch was calculated using the formula for the surface area of a cylinder (mean branch circumference × branch length). The total number of EAB galleries and total number of adult exit holes per branch surface area was the quotient of the total number of galleries, and adult exit holes, and the total surface area ($cm^2$) of the branch, respectively. The percent phloem per branch utilized by EAB larvae was the quotient of the total gallery surface area ($cm^2$) and total surface area ($cm^2$) of the branch. All area measurements are expressed in $m^2$.

***Statistical Analysis.*** Data was analyzed using SigmaStat statistical software (Jandel Scientific, 1992). Percent canopy loss means and standard errors (SEM) were calculated for each study year for each treatment within a given study site for all remaining trees. The Shapiro-Wilk test was used to test for normality and the Levene Median test for equal variance. For each year of data at each study site, a one-way ANOVA was performed to determine if there were any differences among treatment means. If treatment differences were detected within a given year at a given study site, the means were separated at the P = 0.05 level using the Dunn's multiple comparison test. A regression analysis was performed to determine the effect of tree size on the efficacy of an annual 1× basal soil drench of imidacloprid for trees at the Homewood and Skokie study sites. A two-way ANOVA was conducted using the professional Merit 2F and all four Bayer Advanced Tree and Shrub (BATS) treatments at the Riverside, Hinsdale, Naperville, and Woodridge study sites to test for the effects of tree size, tree species, and tree size–tree species interaction for percent canopy loss. All percent canopy loss (PCL) ratings were arcsine transformed before analysis to correct for non-normality and heterogeneity of variance (Jandel Scientific 1992). Real mean percentages are presented in the tables.

## Results

All study trees at all sites were assessed as healthy (mean PCL < 14%) at the beginning of their respective studies. Overall, EAB pressure, as indicated by changes in percent canopy loss, took from two to four years to reach 50% PCL at the green ash study sites. EAB pressure did not develop on the Naperville and Woodridge white ash or the Riverside blue ash trees with no significant differences in PCL for untreated and treated trees. Final mean percent canopy loss for all untreated trees at the Naperville and Woodridge white ash and Riverside blue ash study sites was 22%, 17%, and 12%, respectively (Table 5).

**Skokie Green Ash Study (2007–2011):** During the first three years of the study there were no significant differences in PCL for treated and untreated trees. However, by June, 2011, trees treated with Xytect 75WSP at the 1× rate of 0.56 g a.i./2.54 cm dbh had a significantly higher PCL of 38% than a PCL of less than 19% for trees treated at the 1.5× rate (0.84 g a.i./2.54 cm dbh), and the 2× rate (1.12 g a.i/2.54 cm dbh) of Xytect 75WSP (June, $F = 2.2$; $P = 0.04$) (Table 2). By the end of the study in August, 2011, trees treated at the 1.5× and 2× rates of Xytect 75WSP had significantly lower PCL (less than 21%) compared to the untreated controls (42%) (August, $F = 3.4$; $P < 0.04$). The 1× rate of Xytect 75WSP provided an intermediate level of protection (PCL = 28%) (Table 2).

**Aurora Green Ash Study (2009–2012):** Initial percent canopy loss (PCL) ratings and other general observations indicated EAB pressure was low (PCL less than 7%) on all the Aurora study trees, generally taking approximately two years to build to a level where canopy loss was visually apparent. There were no significant differences among treated trees and the untreated controls in the first two years of the study (PCL less than 24%). However, by June, 2011, significant differences were observed between untreated trees and trees treated with a mid-May, followed by a mid-June, basal soil drench of BATSC and a single mid-May BSD of BATSC2X (June, $F = 2.2$; $P = 0.04$) (Table 3). By August 2011, all treated trees were significantly healthier (PCL of 15% to 31%) (August, $F = 2.8$; $P = 0.03$) compared to untreated trees which were nearly all dead (PCL = 94%) (Table 3). Branch samples taken from untreated trees, during winter, 2011–2012, indicated that EAB pressure at the Aurora site was high and exceeded EAB pressure (i.e. mean number of galleries/$m^2$) for comparable studies by Anulewicz et al. (2008) for heavily infested ash trees suggesting that, in retrospect, the EAB infestation at the Aurora site was probably more developed than originally perceived (Table 8). The percent canopy loss for all treated trees increased by 14% to 16% by June 2012 compared to 2011, possibly in response to the 2012 drought, but then leveled off or decreased by August, 2012 when late summer rains returned. Our findings are consistent with Smitley et al. (2008) who found that PCL increased by approximately 20% following drought periods. All untreated trees were dead (PCL = 100%) by June 2012. By the end of the trial, the mid-May followed by a mid-June basal soil drench of BATSC provided good protection of ash trees (final PCL = 16%). The mid-May followed by a mid-June basal broadcast application of BATSGF1 or a single mid-May basal soil drench application of BATSC2X were not as effective in protecting ash trees (final PCL less than 38%).

**Homewood Green Ash Study (2008–2011):** Three years into the study (June, 2010), significant differences in PCL appeared between treated and untreated trees ($F = 3.2$; $P = 0.03$) (Table 4). All five treatments were highly effective in protecting ash trees from EAB (PCL less than 15%). By 2011, when the study ended, the single mid-May or mid-September basal soil drench

**Table 2. Evaluation of three rates of a single mid-May basal soil drench (BSD) of Xytect 75WSP (imidacloprid 75%) for protection of green ash parkway trees at Skokie, Illinois (SK). Each treatment has 10 single tree replicates.**

| TREATMENT<br>Skokie (SK) green ash parkway trees | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| **Mean dbh = 58 cm (range = 51–89 cm)**<br>**One single mid-May basal soil drench (BSD) of imidacloprid 75%** | | **Mean % canopy loss ratings ± SEM[1]** | | | | | | | |
| | **N[2]** | **Jun 2007** | **Aug 2008** | **Jun 2009** | **Aug 2009** | **June 2010** | **Aug 2010** | **Jun 2011** | **Aug 2011** |
| Xytect 75WSP (0.56 g a.i./2.54 cm dbh) | 10 | 13 ± 2.9a | 19 ± 2.6a | 21 ± 2.8a | 29 ± 3.2a | 37 ± 2.9a | 37 ± 2.9a | 38 ± 1.4b | 28 ± 2.6ab |
| Xytect 75WSP (0.84 g a.i./2.54 cm dbh) | 10 | 1 ± 0.2a | 6 ± 4.4a | 10 ± 2.1a | 11 ± 2.3a | 11 ± 2.3a | 11 ± 2.3a | 10 ± 1.7a | 20 ± 3.1a |
| Xytect 75WSP (1.12 g a.i./2.54 cm dbh) | 10 | 3 ± 0.4a | 7 ± 3.9a | 6 ± 4.4a | 11 ± 2.3a | 17 ± 4.4a | 17 ± 4.4a | 18 ± 1.6a | 20 ± 1.9a |
| UTC | 10 | 12 ± 2.7a | 16 ± 3.7a | 18 ± 4.2a | 23 ± 3.5a | 28 ± 3.7a | 26 ± 3.5a | 21 ± 13.9a | 42 ± 12.1b |
| **Significance:** | | NS[3] | NS | NS | NS | NS | NS | **F=2.2 P=0.04** | **F=3.4 P=0.04** |

[1]Means followed by the same letter are not significantly different (Dunn's test; P<0.05)
[2]N = number of single tree replicates per treatment rate
[3]NS = Not significant (P<0.05)

**Table 3. Evaluation of a single mid-May basal soil drench (BSD) of BATSC (imidacloprid 0.74% + clothianidin 0.37%), a single mid-May basal broadcast application (BBA) of BATSGF1 ((imidacloprid 0.55% + clothianidin 0.275% plus 2-1-1 fertilizer), and a single mid-May BSD of BATSC (imidacloprid 0.74% + clothianidin 0.37%) at the 2X rate for protection of green ash parkway trees at Aurora, Illinois (AU). Each treatment has 10 single tree replications.**

| TREATMENT<br>Aurora (AU) green ash trees | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| **Mean dbh = 43 cm (range = 30-66 cm)** | | **Mean % canopy loss ratings ± SEM[1]** | | | | | | |
| | **N[2]** | **June 2009** | **June 2010** | **August 2010** | **June 2011** | **August 2011** | **June 20120** | **August 2012** |
| BATSC (BSD)<br>(two applications in mid-May and early June) | 10 | 6 ± 2.6a | 10 ± 2.3a | 10 ± 2.3a | 28 ± 4.5a | 15 ± 3.2a | 31 ± 3.3a | 16 ± 2.7a |
| BATSGF1 (BBA)<br>(two applications in mid-May and early June) | 10 | 4 ± 1.6a | 15 ± 2.9a | 15 ± 2.9a | 51 ± 6.1ab | 31 ± 4.7a | 45 ± 4.2a | 37 ± 3.8a |
| BATSC2X (BSD)<br>(one mid-May application at 2X rate) | 10 | 4 ± 1.4a | 19 ± 5.2a | 19 ± 5.2a | 39 ± 6.7a | 22 ± 4.3a | 36 ± 3.9a | 37 ± 3.8a |
| UTC | 10 | 6 ± 2.9a | 23 ± 3.1a | 23 ± 3.1a | 94 ± 1.2b | 94 ± 1.2b | 100 ± 0.0b | 100 ± 0.0b |
| ***Significance:*** | | NS[2] | NS | NS | **F=2.2<br>P=0.04** | **F=2.8<br>P=0.03** | **F=2.4<br>P=0.04** | **F=3.1<br>P<0.001** |

[1]Means followed by the same letter are not significantly different (Dunn's test; P<0.05)
[2]N = number of single tree replicates per treatment rate
[3]NS = Not significant (P<0.05)

**Table 4. Evaluation of a single mid-May and a single mid-September basal soil drench (BSD) of professional Merit 2F (imidacloprid 25%), a single mid-May BSD of BATSC (imidacloprid 0.74% + clothianidin 0.37%), a single mid-May basal broadcast application (BBA) of BATSGF2 (imidacloprid 1.1% plus 2-1-1 fertilizer), or BATSGF3 (imidacloprid 0.55% + clothianidin 0.275% plus 2-1-1 fertilizer) for protection of green ash parkway trees at Homewood, Illinois (HW). Each treatment has 10 single tree replicates.**

| **TREATMENT**<br>**Homewood (HW) green ash trees**<br>**Mean dbh = 41 cm (range = 36-61 cm)**<br>**Mean % Canopy Loss Ratings ± SEM[1]** | **N[2]** | **June 2008** | **June 2009** | **June 2010** | **June 2011** |
|---|---|---|---|---|---|
| Merit 2F (BSD) (one mid-May application) | 10 | 3 ± 2.9a | 12 ± 3.2a | 4 ± 2.6a | 9 ± 4.6a |
| Merit 2F (BSD) (one mid-September application) | 10 | 0 ± 0.0a | 13 ± 2.7a | 9 ± 1.7a | 6 ± 3.0a |
| BATSC (BSD) (one mid-May application) | 10 | 8 ± 5.1a | 21 ± 4.5a | 13 ± 2.6a | 25 ± 2.9ab |
| BATSGF2 (BBA) (one mid-May application) | 10 | 9 ± 1.5a | 7 ± 2.9a | 8 ± 2.1a | 10 ± 3.1a |
| BATSGF3 (BBA) (one mid-May application) | 10 | 6 ± 3.0a | 7 ± 1.5a | 14 ± 1.7a | 12 ± 6.8a |
| UTC | 10 | 11 ± 3.4a | 19 ± 4.2a | 32 ± 4.2b | 80 ± 3.3b |
| ***Significance:*** | | NS[2] | NS | **F=3.2 P=0.03** | **F=33.5 P=0.03** |

[1]Means followed by the same letter are not significantly different (Dunn's test; P<0.05)
[2]N = number of single tree replicates per treatment rate
[3]NS = Not significant (P<0.05)

of professional Merit 2F; the single mid-May basal broadcast application of either BATSGF2 or BATSGF3 were significantly more effective in protecting ash trees (PCL less than 13%) compared to untreated trees ($F = 33.5$; $P = 0.03$). The basal soil drench of BATSC applied annually in mid-May provided intermediate protection (PCL = 25%). 80% of the untreated trees were dead by the end of the study in 2011 (Table 4). In contrast to the Aurora study, good protection of similar size Homewood ash trees was achieved with only a single mid-May basal soil drench or basal broadcast application of imidacloprid alone or in combination with clothianidin. EAB pressure was lower on untreated trees at the Homewood site compared with untreated Aurora trees, and the Homewood study was concluded prior to the onset of the 2012 drought which possibly afforded better protection of the Homewood trees (Table 8). The 25% percent phloem utilization rate for untreated trees at the Homewood site corresponds with low levels of EAB pressure (less than 20%) as defined by Flower et al. (2015).

**Riverside Green Ash Study (2012–2015):** By the second year of the study (June, 2013), significant differences in PCL appeared between all treated and untreated trees with the exception of those trees treated with a single mid-May basal soil drench of professional Merit 2F (June, $F = 8.7$; $P < 0.001$; August, $F = 6.2$; $P < 0.001$) (Table 5). By August, 2013, PCL peaked for all treated trees and reached 57% for untreated trees. During 2014, PCL for all treated trees leveled off and was significantly lower than untreated trees (2014, June, $F = 3.3$; $P < 0.015$ and 2014, August, $F = 32.9$; $P < 0.001$) which were all dead (PCL = 100%). This trend continued through the 2015 growing season with PCL for treated trees remaining below 21%, and significantly different from the untreated trees (2015, June, $F = 31.2$; $P < 0.001$ and 2015, August, $F = 47.8$; $P < 0.001$) (Table 5). All untreated trees were dead by the end of the study in 2015.

**Hinsdale Green Ash Study (2012–2015):** Significant differences in PCL were

first observed in August, 2014 between untreated trees ($F = 11.4$; $P = 0.04$) and trees treated with either a single mid-May basal soil drench of professional Merit 2F, or a single mid-May basal broadcast application of the homeowner formulation of BATSGF1 (Table 5). Annual mid-May basal soil drench treatments of homeowner formulations of BATSC and BATSCF, and a basal broadcast application of the homeowner formulation of BATSFG4, were not as effective in protecting green ash trees. By August, 2015, when the study ended, the percent canopy loss for trees treated with a basal soil drench of professional Merit 2F, or a basal broadcast application of either BATSGF1 or BATSGF4 provided significantly better protection of ash trees compared to the untreated trees. Trees treated with basal soil drenches of BATSC or BATSCF provided intermediate protection of Hinsdale green ash trees. Percent canopy loss for untreated trees approached 60% by the end of the study (Table 5).

**Riverside Blue Ash Study (2012–2015):** All of the blue ash study trees were in excellent condition at the beginning of the study (PCL less than 12%). EAB pressure failed to build with PCL on untreated control trees less than 13% after four years. There was no significant difference in PCL between treated and untreated blue ash trees (Table 5). The low PCL associated with the untreated Riverside blue ash trees is consistent with findings by Tanis and McCullough (2012) where they found a higher survival rate of blue ash following an EAB infestation. Unprotected green ash trees in the immediate area around the study site were dying or dead from EAB.

**Naperville and Woodridge White Ash Studies (2012–2015):** All Naperville and Woodridge white ash study trees were very healthy at the beginning of the study in 2012, with PCLs less than 16% and 10%, respectively. EAB pressure failed to build throughout the study period, at both sites, as indicated by PCL of 22% and 17% for all untreated study trees, respectively. There were no significant differences in PCL between treated and untreated trees at either site over the four-year period (Table 5). Unprotected green ash trees in and around both study sites were dying or dead from EAB.

**Glen Ellyn Green Ash Study (2010–2014):** EAB pressure was slow to build from June, 2010 to June, 2013. Beginning in June and through August, 2013, significant differences in PCL occurred between trees treated with an annual BSD of Arena 50WDG (PCL = 13%) and the untreated controls (June, $F = 2.4$; $P = 0.04$ and August, $F = 2.3$; $P = 0.04$) (Table 6). The remaining treated trees had a slightly higher PCL of 15% to 19%. By August, 2014, all treated trees had significantly lower PCL (less than 23%) compared to untreated trees, which were all dead (PCL = 100%) (June, $F = 3.9$; $P < 0.001$ and August, $F = 8.3$; $P < 0.001$). Specifically, trees treated with an annual mid-June basal soil drench of Arena 50WDG, Arena 50WDG + Safari 20SG, an annual mid-June basal trunk spray of Arena 50WDG, or an annual mid-June basal broadcast application of Safari 2G, had significantly lower PCL (mean = 13%) compared to trees treated with a mid-June or mid-July basal soil drench of Safari 20SG (mean PCL = 20%) (Table 6).

**Fermi Lab Village Green Ash Study (2008–2015):** Our EAB trap tree monitoring program, implemented in May, 2008, indicated it took approximately three years for the EAB infestation to spread to the Fermi Lab Village study site, a distance of approximately 1.8 km. These observations are consistent with the rate of natural spread of EAB (Herms and McCullough 2014). PCL from 2008 to 2010 did not exceed 15% for all treated and untreated trees (Table 7). The 2011 field season appeared to be the tipping point, and coincided with when EAB was first detected in trap trees along the western edge of the study site. Beginning with the 2011 field season, treated trees had significantly lower PCL (mean less than 17%) compared to untreated control trees (PCL = 30%) ($F = 2.5$; $P < 0.02$) (Table 7). From 2012 until the end of the study in 2015, PCL for all treated trees declined and remained below 6%, while PCL for unprotected trees increased to 90% by 2013. All untreated trees were dead by 2015 (2012, $F = 4.6$; $P < 0.001$; 2013, $F = 51.6$; $P < 0.001$; 2014, $F = 52.4$; $P < 0.001$; 2015, $F = 49.2$; $P < 0.001$) (Table 7). In addition, numerous untreated, non-study trees growing in areas around the FLV site, were also dead.

**Role of ash tree size in the efficacy of an imidacloprid basal soil drench.** To evaluate the role of tree size, for treatment efficacy of a 1× rate of an annual mid-May imidacloprid basal soil drench application, treated and untreated green ash trees, at the Skokie and Homewood study sites were grouped separately for a regression analysis because both studies were started in 2007 and 2008, respectively; and the trees were in a similar condition (PCL equal to 7% and 6%, respectively) at the beginning of their respective studies. Regression analysis revealed that tree size had no significant effect on rates of decline for either untreated control trees ($F = 0.01$, $R^2 = 0.01$, $P = 0.95$) or trees treated with a 1× rate of an annual mid-May imidacloprid basal soil drench ($F = 1.71$, $R^2 = 0.10$, $P = 0.21$). Both smaller (less than 50 cm dbh) and larger (greater than 50 cm dbh) untreated control trees at the Homewood and Skokie study sites declined to a

**Table 5. Evaluation of a single mid-May basal soil drench (BSD) of professional Merit 2F (imidacloprid 25%), or BATSC (imidacloprid 0.74% + clothianidin 0.37%), a single mid-May basal broadcast application (BBA) of BATSGF3 ((imidacloprid 0.55% + clothianidin 0.275% plus 2-1-1 fertilizer) or BATSGF4 (imidacloprid 1.1% plus 2-1-1 fertilizer), and a single mid-May BSD of BATSCF (imidacloprid 1.47% plus 2-1-1 fertilizer) all at the 2X rate for protection of green, white, and blue ash park and parkway trees at Riverside (RV), Hinsdale (HI), Naperville (NV), and Woodridge (WR), Illinois. Each treatment has 10 single tree replicates for the Riverside and Hinsdale green ash sites, five single tree replicates each for the Naperville and Woodridge white ash sites, and six single tree replicates for the Riverside blue ash study site.**

| TREATMENT<br>One mid-May BSD or BBA at 2X application rate | | Mean % canopy loss ratings ± SEM[1] | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | 2012 | | 2013 | | 2014 | | 2015 | |
| | N[2] | June | August | June | August | June | August | June | August |
| **Riverside (RV) green ash trees**<br>**Mean dbh = 61 cm (range = 51–91 cm)** | | | | | | | | | |
| Merit 2F (BSD) | 10 | 20 ± 4.0a | 24 ± 4.2a | 20 ± 2.4ab | 40 ± 6.9ab | 32 ± 2.1a | 13 ± 1.7a | 30 ± 13.6a | 18 ± 6.0a |
| BATSC (BSD) | 10 | 25 ± 5.7a | 25 ± 5.7a | 15 ± 2.3a | 35 ± 6.9ab | 32 ± 2.5a | 22 ± 1.7a | 20 ± 13.1a | 20 ± 5.7a |
| BATSGF1 (BBA) | 10 | 19 ± 4.6a | 24 ± 4.2a | 14 ± 1.2a | 33 ± 4.3ab | 33 ± 6.9a | 29 ± 7.1a | 21 ± 8.4a | 15 ± 2.2a |
| BATSGF4 (BBA) | 10 | 20 ± 4.5a | 17 ± 3.9a | 14 ± 2.5a | 26 ± 4.2a | 31 ± 7.1a | 20 ± 6.8a | 10 ± 13.2a | 20 ± 7.6a |
| BATSCF (BSD) | 10 | 20 ± 3.8a | 19 ± 3.7a | 14 ± 2.5a | 29 ± 4.4a | 26 ± 8.4a | 28 ± 8.5a | 12 ± 9.7a | 19 ± 3.3a |
| UTC | 10 | 13 ± 5.5a | 13 ± 5.5a | 27 ± 4.6b | 57 ± 9.5b | 72 ± 4.4b | 100 ± 0.0b | 100 ± 0.0b | 100 ± 0.0b |
| ***Significance:*** | | NS[2] | NS | **F=8.7**<br>**P<0.001** | **F=6.2**<br>**P<0.001** | **F=3.3**<br>**P=0.015** | **F=32.9**<br>**P<0.001** | **F=31.2**<br>**P<0.001** | **F=47.8**<br>**P<0.001** |
| **Hinsdale (HI) green ash trees**<br>**Mean dbh = 51 cm (range = 46–91 cm)** | | | | | | | | | |
| Merit 2F (BSD) | 10 | 13 ± 2.1a | 14 ± 2.0a | 24 ± 4.0a | 34 ± 2.4a | 27 ± 5.6a | 30 ± 5.8a | 29 ± 3.5a | 27 ± 3.1a |
| BATSC (BSD) | 10 | 12 ± 1.7a | 14 ± 1.9a | 30 ± 5.7a | 31 ± 4.9a | 32 ± 7.1a | 34 ± 7.8ab | 37 ± 3.6ab | 37 ± 3.6ab |
| BATSGF1 (BBA) | 10 | 14 ± 2.2a | 13 ± 2.1a | 26 ± 4.6a | 28 ± 4.4a | 29 ± 7.8a | 31 ± 8.6a | 27 ± 2.1a | 31 ± 1.9a |
| BATSGF4 (BBA) | 10 | 22 ± 2.9a | 23 ± 2.9a | 33 ± 4.6a | 38 ± 3.7a | 34 ± 4.5a | 39 ± 6.8ab | 30 ± 3.5a | 30 ± 2.7a |
| BATSCF (BSD) | 10 | 12 ± 1.7a | 16 ± 2.1a | 28 ± 3.9a | 29 ± 3.9a | 37 ± 4.8a | 39 ± 5.9ab | 39 ± 6.6ab | 41 ± 6.9ab |
| UTC | 10 | 22 ± 2.9a | 25 ± 3.1a | 39 ± 10.1a | 45 ± 5.5a | 43 ± 6.2a | 59 ± 7.8b | 59 ± 11.9b | 59 ± 11.9b |
| ***Significance:*** | | NS[2] | NS | **NS** | **NS** | **NS**<br>**P=0.04** | **F=11.4**<br>**P=0.03** | **F=3.0**<br>**P=0.03** | **F=3.3** |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| **Riverside (RV) blue ash trees**<br>**Mean dbh = 26 cm (range = 19–36 cm)** | | | | | | | | | |
| Merit 2F (BSD) | 6 | 9 ± 0.7a | 6 ± 0.7a | 4 ± 0.0a | 4 ± 1.0a | 3 ± 0.0a | 4 ± 0.0a | 7 ± 1.4a | 7 ± 1.4a |
| BATSC (BSD) | 6 | 10 ± 1.1a | 8 ± 1.1a | 7 ± 1.8a | 9 ± 1.9a | 3 ± 0.0a | 4 ± 1.2a | 9 ± 1.5a | 9 ± 1.5a |
| BATSGF1 (BBA) | 6 | 9 ± 1.1a | 5 ± 1.1a | 7 ± 1.1a | 6 ± 0.7a | 3 ± 0.7a | 4 ± 0.8a | 8 ± 1.45a | 8 ± 1.4a |
| BATSGF4 (BBA) | 6 | 8 ± 1.0a | 2 ± 1.0a | 5 ± 1.0a | 6 ± 0.7a | 2 ± 0.7a | 3 ± 0.2a | 6 ± 1.2a | 6 ± 1.2a |
| BATSCF (BSD) | 6 | 9 ± 0.0a | 9 ± 0.0a | 9 ± 0.7a | 10 ± 0.8a | 6 ± 0.7a | 7 ± 0.8a | 8 ± 1.7a | 8 ± 1.7a |
| UTC | 6 | 11 ± 2.0a | 7 ± 1.1a | 5 ± 0.7a | 6 ± 0.7a | 7 ± 1.0a | 7 ± 0.8a | 12 ± 2.2a | 12 ± 2.2a |
| ***Significance:*** | | **NS[2]** | **NS** | **NS** | **NS** | **NS** | **NS** | **NS** | **NS** |
| **Naperville (NV) white ash trees**<br>**Mean dbh = 58 cm (range = 51–64 cm)** | | | | | | | | | |
| Merit 2F (BSD) | 5 | 4 ± 3.0a | 4 ± 3.0a | 8 ± 2.0a | 9 ± 2.1a | 17 ± 4.9a | 16 ± 3.9a | 19 ± 4.3a | 19 ± 4.3a |
| BATSC (BSD) | 5 | 6 ± 2.6a | 7 ± 2.5a | 10 ± 3.1a | 9 ± 1.5a | 8 ± 2.5a | 10 ± 0.0a | 23 ± 8.0a | 23 ± 8.0a |
| BATSGF1 (BBA) | 5 | 3 ± 3.1a | 3 ± 3.1a | 10 ± 3.1a | 13 ± 3.6a | 5 ± 4.0a | 6 ± 3.9a | 20 ± 0.0a | 20 ± 0.0a |
| BATSGF4 (BBA) | 5 | 9 ± 3.3a | 9 ± 3.3a | 21 ± 4.1a | 18 ± 2.8a | 10 ± 0.0a | 15 ± 0.5a | 17 ± 3.3a | 17 ± 3.3a |
| BATSCF (BSD) | 5 | 9 ± 1.5a | 8 ± 1.7a | 11 ± 3.3a | 15 ± 3.5a | 12 ± 5.0a | 17 ± 4.2a | 28 ± 4.8a | 28 ± 4.8a |
| UTC | 5 | 15 ± 5.0a | 18 ± 5.1a | 12 ± 1.7a | 15 ± 5.0a | 12 ± 2.4a | 17 ± 3.6a | 22 ± 8.0a | 22 ± 8.0a |
| ***Significance:*** | | **NS[2]** | **NS** | **NS** | **NS** | **NS** | **NS** | **NS** | **NS** |
| **Woodridge (WR) white ash trees**<br>**Mean dbh = 37 cm (range = 25–48 cm)** | | | | | | | | | |
| Merit 2F (BSD) | 5 | 6 ± 0.7a | 6 ± 0.7a | 7 ± 2.0a | 9 ± 1.0a | 7 ± 2.0a | 10 ± 0.0a | 15 ± 1.4a | 15 ± 1.4a |
| BATSC (BSD) | 5 | 9 ± 1.1a | 9 ± 1.1a | 8 ± 1.9a | 8 ± 1.9a | 5 ± 0.0a | 9 ± 1.2a | 14 ± 2.5a | 14 ± 2.5a |
| BATSGF1 (BBA) | 5 | 9 ± 1.1a | 9 ± 1.1a | 9 ± 1.1a | 12 ± 1.7a | 6 ± 0.7a | 7 ± 0.8a | 11 ± 2.5a | 11 ± 2.5a |
| BATSGF4 (BBA) | 5 | 8 ± 1.0a | 8 ± 1.0a | 8 ± 1.9a | 6 ± 0.7a | 6 ± 0.7a | 9 ± 1.2a | 12 ± 7.2a | 12 ± 7.2a |
| BATSCF (BSD) | 5 | 10 ± 0.0a | 10 ± 0.0a | 6 ± 0.7a | 7 ± 0.8a | 6 ± 0.7a | 7 ± 0.8a | 12 ± 1.7a | 12 ± 1.7a |
| UTC | 5 | 7 ± 2.0a | 7 ± 2.0a | 6 ± 0.7a | 6 ± 0.7a | 7 ± 2.0a | 7 ± 0.8a | 17 ± 2.2a | 17 ± 2.2a |
| Significance: | | | | **NS[2]** | **NS** | **NS** | **NS** | **NS** | **NS** | **NS** | **NS** |

[1]Means followed by the same letter are not significantly different (Dunn's test; P<0.05)
[2]N = number of single tree replicates per treatment rate
[3]NS = Not significant (P<0.05)

**Table 6. Evaluation of a single mid-June basal soil drench (BSD) of Arena 50WDG (clothianidin 50%), Safari 20SG (dinotefuran 20%), or Safari 20SG + Arena 50WDG (dinotefuran 20% + clothianidin 50%), a single mid-July BSD of Safari 20SG (dinotefuran 20%), a single mid-June basal trunk spray (BTS) of Arena 50WDG (clothianidin 50%) or Safari 20SG (dinotefuran 20%), and a single mid-June basal broadcast application (BBA) of Safari 2G (dinotefuran 2%) for protection of green ash parkway trees at Glen Ellyn, Illinois (GE). Each treatment has seven single tree replicates.**

| **TREATMENT**<br>**Glen Ellyn (GE) green ash parkway trees**<br>**Mean dbh = 41 cm (range = 28-48 cm)** | | **Mean % Canopy Loss Ratings ± SEM[1]** | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | **N[2]** | **June 2010** | **June 2011** | **August 2011** | **June 2012** | **August 2012** | **June 2013** | **August 2013** | **June 2014** | **August 2014** |
| Arena 50WDG (BSD) | 7 | 4 ± 2.3a | 6 ± 1.5a | 12 ± 2.9a | 5 ± 2.1a | 12 ± 3.1a | 13 ± 3.1a | 12 ± 3.2a | 13 ± 3.1a | 13 ± 3.1a |
| Safari 20SG (BSD) (*mid-June application*) | 7 | 5 ± 0.9a | 20 ± 4.3a | 17 ± 8.2a | 12 ± 4.9a | 27 ± 13.2a | 19 ± 3.9ab | 16 ± 4.0ab | 20 ± 5.9a | 18 ± 5.6b |
| Safari 20SG + Arena 50WDG (BSD) | 7 | 6 ± 1.5a | 11 ± 4.6a | 18 ± 7.9a | 10 ± 4.2a | 13 ± 3.0a | 16 ± 2.6ab | 15 ± 0.0ab | 17 ± 2.9a | 16 ± 2.8a |
| Safari 20SG (BSD) (mid-July application) | 7 | 6 ± 1.4a | 6 ± 1.7a | 13 ± 3.1a | 17 ± 8.2a | 21 ± 5.4a | 17 ± 4.9ab | 16 ± 2.6ab | 18 ± 5.4a | 22 ± 6.1b |
| Arena 50WDG (BTS) | 7 | 5 ± 2.2a | 7 ± 2.1a | 11 ± 4.9a | 6 ± 2.8a | 16 ± 2.5a | 16 ± 3.7ab | 16 ± 3.7ab | 17 ± 2.0a | 10 ± 0.0a |
| Safari 20SG (BTS) | 7 | 4 ± 1.7a | 11 ± 4.9a | 16 ± 2.5a | 8 ± 2.6a | 14 ± 3.0a | 16 ± 2.8ab | 16 ± 2.8ab | 17 ± 2.4a | 11 ± 4.9a |
| Safari 2G (BBA) | 7 | 4 ± 3.8a | 14 ± 3.3a | 20 ± 4.3a | 7 ± 4.1a | 20 ± 4.6a | 19 ± 3.9ab | 17 ± 3.6ab | 20 ± 4.7a | 16 ± 2.7a |
| UTC | 7 | 4 ± 2.4a | 11 ± 4.6a | 14 ± 3.2a | 20 ± 10.1a | 34 ± 12.1a | 43 ± 12.5b | 43 ± 12.5b | 100 ± 0.0b | 100 ± 0.0c |
| ***Significance:*** | | **NS[3]** | **NS** | **NS** | **NS** | **NS** | **F=2.4 P=0.04** | **F=2.30 P=0.04** | **F=3.89 P<0.001** | **F=8.25 P<0.001** |

[1]Means followed by the same letter are not significantly different (Dunn's test; P<0.05)
[2]N = number of single tree replicates per treatment rate
[3]NS = Not significant (P<0.05)

**Table 7. Evaluation of a single mid-May trunk injection (TI) of Tree-age (emamectin benzoate 4%) for protection of green ash park and parkway trees at the Fermi Lab Village (FLV), Fermi National Accelerator Laboratory (FNAL), Batavia, Illinois. Each treatment has 10 single tree replicates.**

**TREATMENT**
**Fermi Lab Village (FVL) green ash park and parkway trees** **Mean % Canopy Loss Ratings ± SEM[1]**
**Mean dbh=44 cm (range=38-50) for small trees**
**Mean dbh=60 cm (range 50-70 cm) for large trees**
**One single mid-May trunk injection of 4% emamectin benzoate (EB) in 2008 and again in mid-September, 2012**

| | N[2] | 2008 | 2009 | 2010 | 2011 | 2012[1] | 2013 | 2014 | 2015 |
|---|---|---|---|---|---|---|---|---|---|
| Tree-Age Quik-Jet-Small Trees (0.2 g a.i./2.54 cm dbh) | 10 | 7 ± 1.9a | 11 ± 3.1a | 12 ± 3.1a | 15 ± 0.0a | 5 ± 0.0a | 5 ± 0.0a | 5 ± 0.0a | 5 ± 0.0a |
| Tree-Age Tree IV-Small Trees (0.3 g a.i./2.54 cm dbh) | 10 | 12 ± 1.1a | 7 ± 0.8a | 14 ± 2.9a | 16 ± 2.8a | 5 ± 0.0a | 5 ± 0.0a | 5 ± 0.0a | 5 ± 0.0a |
| Tree-Age Quik-Jet-Large Trees (0.4 g a.i./2.54 cm dbh) | 10 | 8 ± 1.8a | 14 ± 2.5a | 12 ± 2.7a | 15 ± 0.0a | 5 ± 0.0a | 5 ± 0.0a | 5 ± 0.0a | 5 ± 0.0a |
| Tree-Age Tree IV-Large Trees (0.6 g a.i./2.54 cm dbh) | 10 | 6 ± 1.8a | 14 ± 3.5a | 6 ± 1.3a | 9 ± 1.9a | 5 ± 0.0a | 5 ± 0.0a | 5 ± 0.0a | 5 ± 0.0a |
| UTC | 10 | 11 ± 5.5a | 14 ± 4.1a | 11 ± 2.2a | 30 ± 0.0b | 56 ± 5.7b | 90 ± 3.6b | 95 ± 3.2b | 100 ± 0.0b |
| ***Significance:*** | | **NS[2]** | **NS** | **NS** | **F=2.5 P<0.02** | **F=4.6 P<0.001** | **F=51.6 P<0.001** | **F=52.4 P<0.001** | **F=49.2 P<0.001** |

[1]Means followed by the same letter are not significantly different (Dunn's test; P<0.05)
[2]N = number of single tree replicates per treatment rate
[3]NS = Not significant (P<0.05)

**Table 8. Summary of percent phloem area utilized by EAB larvae, mean number of galleries/m², and mean number of adult exit holes/m² for branch samples taken from treated and untreated trees at the Aurora and Homewood study sites during the winter of 2011–2012.**

| BRANCH SAMPLING VARIABLE[1] | STUDY SITE | | | | |
|---|---|---|---|---|---|
| | **N** | **Aurora[2]** | **N** | **Homewood[3]** | **Significance** |
| Mean % phloem area utilized for untreated trees | 40 | 61 ± 6.2b | 44 | 25 ± 3.0a | t = 5.6, P=0.005 |
| Mean number of galleries/m² for untreated trees | 40 | 268 ± 31.4b | 44 | 64 ± 3.5a | t = 6.5, P=0.003 |
| Mean number of exit holes/m² for untreated trees | 40 | 179 ± 6.9b | 44 | 16 ± 2.3a | t = 6.0, P=0.004 |

[1]Means followed by the same letter across rows are not significantly different (t-test, P<0.05)
[2]N equals a total of 40 branch samples taken for all remaining untreated trees
[3]N equals a total 44 branch samples taken for all remaining untreated trees

weakened condition and an unacceptable appearance (80% and 42%, respectively). Treated trees, at both sites, remained in excellent to good condition with 9% and 20% percent canopy loss for Homewood and Skokie trees, respectively (Tables 2 and 4).

**Role of ash tree size, tree species, and tree size-species interaction on the efficacy of basal soil drenches of imidacloprid applied alone or in combination with clothianidin.** A two-way ANOVA procedure was used to examine the role of tree size, species, and their interaction on the efficacy of all trees treated with an annual mid-May 2× application of professional Merit 2F and Bayer Advanced Tree and Shrub homeowner products (imidacloprid alone, and imidacloprid plus clothianidin) (refer to Table 1) for the smaller white ash (Woodridge), larger white ash (Naperville), larger green ash (Riverside, Hinsdale), and small blue ash (Riverside) trees. The white, green, and blue ash trees from the Woodridge, Riverside, Naperville, and Hinsdale study sites were grouped together because all four studies were initiated in 2012 were of same duration (2012–2015), and all the trees were in good to excellent condition (10 to 20% canopy loss) at the beginning of their respective studies. Results from the two-way ANOVA revealed that ash tree size ($F = 1.2$; $P = 0.30$), ash tree species ($F = 0.62$; $P = 0.43$), and, tree size- species interaction ($F = 0.9$; $P = 0.36$) were not significant for percent canopy loss.

**Phloem utilization by EAB larvae at the Aurora, Homewood, and Fermi Lab Village Sites.** A summary of phloem utilization by EAB larvae, mean number of total galleries per m², and mean number of exit holes per m² for untreated trees at the Aurora and Homewood study sites is presented in Table 8. Percent phloem utilization of remaining untreated Aurora green ash trees, by EAB larvae, was significantly greater ($T = 5.6$; $P = 0.005$), by over two times, compared with the Homewood site (61% versus 25%). Over four times as many galleries were constructed, per m² of branch surface area, on untreated trees at the Aurora site (268 versus 64 galleries) compared with untreated Homewood trees ($T = 6.5$; $P = 0.003$), and over 10 times as many exit holes per m² of branch surface area were counted on untreated Aurora study trees compared with untreated Homewood trees (179 versus 16 exit holes) ($T = 6.0$, $P = 0.004$). A regression comparing percent canopy loss with the number of branch galleries and adult EAB exit holes per m² of branch surface area, revealed a very strong relationship between percent canopy loss and the number of galleries/m² ($R^2 = 0.90$; $P < 0.001$) and adult EAB exit holes/m² ($R^2 = 0.88$; $P = 0.002$) accounting for 90% and 88% of the variation, respectively suggesting that EAB pressure was much higher at the Aurora site compared with the Homewood site by the end of the 2011 growing season.

Branch samples taken from remaining untreated trees at the Skokie site revealed 7% phloem utilization by EAB larvae, a mean of 24 larval galleries/m², and a mean of 10 adult exit holes per m² suggesting that EAB pressure was low and was very similar to the Homewood site.

While only one treated and two untreated study trees were sampled at the Fermi Lab Village site, the differences in percent phloem utilization, mean number of

galleries/m$^2$, and mean number of exit holes/m$^2$ between treated and untreated trees were very apparent. Branch samples taken from the single tree treated with emamectin benzoate had 0% phloem utilized, no EAB larval galleries, and no adult EAB exit holes. In contrast, the two untreated trees had a mean larval phloem utilization of 50%, 130 galleries/m$^2$, and 90 exit holes/m$^2$.

## Discussion

Neonicotinoid-class insecticides have been shown to be effective when applied as basal soil drenches and/or via soil injections for wood-boring and tunneling insect pests, and these products are available to homeowners as well as professional applicators, in part, due to their systemic action, and having shown effectiveness in protecting ash trees less than 38 cm dbh from EAB (Wang et al. 2005; Smitley et al. 2010 a,b, 2015; McCullough et al. 2011; Herms et al. 2014). However, only a limited number of studies have examined the efficacy of neonicotinoids for protection of ash trees larger than 38 cm dbh, and more specifically ash trees over 50 cm dbh (Smitley et al. 2010a, b, 2015; Bick et al. 2018; McCullough et al. 2019). In an effort to provide homeowners and professional practitioners with options for chemically protecting larger green and white ash trees from EAB, we evaluated various active ingredients and their combinations, at different rates, formulations, application methods and timing of neonicotinoid-class insecticides, and emamectin benzoate.

The authors recognize that PCL is a relative measure of insecticide efficacy, but in this study the very strong correlation between the number of galleries/m$^2$ and percent canopy loss supports the use of PCL as a reliable measure of insecticide efficacy. In addition, Flower et al. (2015) found the ash canopy condition rating system to be a proxy of EAB densities at the tree level, and that the canopy rating system was able to identify trees in the early stages of an EAB infestation with relatively low levels of EAB (less than 20% gallery cover or less than 40 EAB/m$^2$). Further, visual estimates of PCL are used in field studies to evaluate insecticide performance, phytotoxicity, and plant damage caused by wood-boring insect pests of woody plants (Ball and Simmons 1980; Anulewicz et al. 2007, 2008; Smitley et al. 2008, 2010 a,b, 2015; McCullough et al. 2011; Nielsen et al. 2011; Bick et al. 2018; Subburayalu and Syndor 2018).

There are many factors that may affect the efficacy and use of an insecticide for protecting ash trees from EAB, including but not limited to, EAB pressure, timing and method of application, overall tree health, related ash tree insect pests and diseases, soil moisture, and ash tree species composition. Here, we will briefly discuss the potential effects of EAB population pressure, ash species composition, and drought may have on the efficacy of chemical treatments, and the importance of recognizing and adjusting for these factors when formulating and implementing a comprehensive EAB management plan.

### Efficacy of imidacloprid applied alone and imidacloprid in combination with clothianidin for the protection of ash trees from EAB

**Smaller ash trees (less than 50 cm dbh).** Collectively, all commercial and homeowner formulations, rates, and application timing of imidacloprid used alone or in combination with clothianidin provided good to excellent protection (PCL less than 17%) of Aurora and Homewood green ash, Woodridge white ash, and Riverside blue ash parkway trees. Further, there does not appear to be any added benefit to applying imidacloprid combined with clothianidin. Our findings are consistent with previous studies by McCullough et al. (2011), Tanis and McCullough (2012), Smitley et al. (2015), Bick et al. (2018) for trees with a similar dbh, and treated with similar active ingredients, combinations, timing, and rates of application. The lower level of protection of the basal broadcast application of BATSGF1 and basal soil drench of BATSC2X treatments at the Aurora study site is not clear. It is possible that the active ingredient of the granular formulation (i.e. BATSGF1) did not thoroughly leach from the granules, but was not investigated in this study. Another possible explanation could be EAB pressure. The much higher EAB pressure at the Aurora site, compared with the Homewood site, in conjunction with the 2012 drought may be partially responsible for the reduced protection of the basal broadcast application of BATSGF1 and basal soil drench of BATSC2X treatments at the Aurora study site. Additionally, the Homewood study was concluded, prior to the onset of the 2012 drought. These findings illustrate the importance of the role of soil moisture conditions in the uptake and distribution of protective chemicals, and of applying these chemical treatments well in advance of a building EAB infestation while trees are still healthy and before damage is very apparent. Failure to act can result in lack of effective EAB control and extensive loss of tree cover (Herms et al. 2019).

**Larger ash trees (greater than 50 cm dbh).** Taken together**,** all commercial and homeowner formulations, rates, application methods, and timing of imidacloprid

used alone or in combination with clothianidin, applied annually at the 2× rate provided good to excellent protection of green and white ash park and parkway trees at the Riverside Hinsdale, Naperville, and Skokie study sites for trees with a mean dbh greater than 50 cm dbh; the only exception being the Hinsdale trees treated in mid-May at the 2× rate with basal soil drenches of BATSC and BATSCF. Interestingly, in this study, tree size (dbh) did not have any significant effect on the rate of decline of Homewood and Skokie untreated trees ($R^2 = 0.01$, $P = 0.95$) or trees treated ($R^2 = 0.10$, $P = 0.21$) with an annual 1× basal soil drench of imidacloprid. However, efficacy of the 1× imidacloprid basal soil drench did decrease for the larger treated Skokie trees (final PCL = 28%) probably because of the relationship between tree size (dbh), tree surface area, and phloem biomass ((LeGoff and Ottorini 1996, McCullough and Siegert 2007, Smitley et al. 2010b). Our findings are in contrast to a study by Smitley et al. (2010b), where it was found that canopy thinning was dependent on trees size (dbh) ($R^2 = 0.48$, $P < 0.002$) for trees receiving an annual basal soil drench of imidacloprid at the same rate and timing as in our study, and treated trees over 38 cm dbh declined to a weakened state and undesirable appearance (PCL of 35 to 80%) by the end of their study. In our study, both the smaller Homewood (mean dbh = 41 cm) and larger (mean dbh = 58 cm) Skokie treated trees remained in excellent to good condition, respectively. It is possible that the lower EAB pressure at the Skokie site may have afforded the larger trees the ability to fight off EAB allowing the 1× imidacloprid rate to still provide some level of EAB protection. These findings point to the need and importance of increasing treatment rates for larger trees, and illustrates the effect EAB pressure can have on insecticide treatment efficacy for both small and large trees. The reduced level of protection of the mid-May basal soil drenches of BATSC and BATSCF for the Hinsdale green ash trees is also unclear, but may be partially explained by the regional record-setting drought beginning in fall, 2011 and continuing through late summer, 2012. The overall 20% increase in percent canopy loss of treated green ash trees at the Hinsdale site in June, 2013, immediately following the 2012 drought, is consistent with studies by Smitley et al. (2015) which found an increase of 5% to 35% canopy loss following a 2008 spring and summer drought. A similar but delayed percent canopy loss drought response was observed by August, 2013 for similar sized Riverside green ash trees treated with the same products, application methods and timing. Local field observations by the senior author during the 2012 and mid to late 2013 growing seasons revealed common landscape and parkway tree species going into early fall color and leaf scorch along with premature leaf drop all indicating tree stress conditions suggesting that the failure of the basal soil drenches of BATSC and BATSCF, to protect the larger Hinsdale study trees, was probably due more to drought conditions than EAB pressure.

Further, there was also a differential tree recovery response following the 2012 drought between treated trees at both the Hinsdale and Riverside sites. While not investigated in this study, this differential response may be partially due to differences in available soil rooting volume, degree of precipitation runoff and/or infiltration, and related soil moisture levels between the two sites. The Riverside green ash trees were growing in a park setting in the Des Plaines River floodplain with better drained undisturbed soils, in contrast to the Hinsdale trees which were growing in an older residential area with narrow parkways containing typical compacted urban soils with limited soil volume. There was less local precipitation in May, 2012, coming in 11-day period, compared with 2.54 cm more rainfall in June, 2012, but in only four days (Illinois State Water Survey Climate Data, 2012). It is possible that less infiltration and more runoff may have occurred for trees growing in dry crusted soil conditions in the Hinsdale residential parkways compared with Riverside trees growing in a park setting with greater infiltration, less compaction, and less runoff, which may have contributed to greater plant stress reducing the trees ability to fight off the EAB (Larsson 1989, Craul 1999, Herms 2002, Huberty and Denno 2004, Gregory and Dukes 2006, Fahey et al. 2013). Cregg and Dix (2001) found that green ash trees planted in a downtown urban area experienced more drought stress and suffered higher damage from clearwing borers than did trees in a park-like campus. With the return of above-normal precipitation during the 2014 and 2015 growing seasons (Illinois State Water Survey, September 2014 and 2015), treated Riverside ash trees began to recover, but treated Hinsdale trees failed to recover. These aforementioned abiotic factors (i.e. soil conditions, rooting volume, precipitation, and water infiltration) may have affected the uptake and subsequent distribution of the insecticide treatments, resulting in a lower level of protection for the Hinsdale treated trees.

**Susceptibility of ash species to EAB.** All North American ash species are considered susceptible to EAB, but green ash and black ash (*F. nigra*) appear to be more highly preferred, followed by white ash

and blue ash (Herms et al. 2019). Previous studies and field observations by the authors and tree care professionals all have indicated that EAB infestations and subsequent green ash tree mortality appears to progress more rapidly compared to white and blue ash (Anulewicz et al. 2007, 2008; Rebek et al. 2008; Tanis and McCullough 2012, 2015; Herms et al. 2014; Herms 2015; Robinette and McCullough 2019; Miller, F. unpublished). Interestingly, over a four-year study period, the PCL of untreated white ash trees at the Naperville, and Woodridge study sites, and untreated Riverside blue ash trees never exceeded 23%, 18%, and 13%, respectively even though adjacent unprotected, non-study green ash trees at all three study sites were dead or dying. However, in our study tree size, species and their interaction did not explain differences in percent canopy loss by treatments at these study sites. These apparent differences in host susceptibility, and the rate and degree of mortality among North American ash species are not clearly understood, and probably include mechanical and chemical host plant characteristics. For example, differences in EAB host susceptibility may be related to differences in host volatiles, nutritional, and defensive compounds (Eyles et al. 2007; Chen and Poland 2009, 2010; Chen et al. 2011, 2012; Martinson et al. 2014; Herms 2015; Poland et al. 2015; Showalter et al. 2018). Additionally, initial and building EAB pressure, combining protective insecticides with ash tree population reduction and sanitation (i.e. "culling the herd" and tree removals) (McCullough and Mercader 2012, Lewis and Turcotte 2015, Rutkowski and Mitz 2017, McCullough et al. 2019) and biological control (Loerch and Cameron 1984, Anulewicz et al. 2008, Dirr 2009, Marshall et al. 2013, Duan et al. 2014, Bauer et al. 2015, Wang et al. 2016, Miller and Gould 2018) may also be contributing factors affecting ash EAB susceptibility and survival. Regardless of the factor(s) responsible for this differential mortality rate observed between green, white, and blue ash trees, the apparently less susceptible white and blue ash to EAB have the potential to impact EAB management decision-making processes, particularly for communities late to apply protective chemical treatments or with limited tree care budgets. Given a choice, some tree care managers are opting to protect existing white and blue ash trees and their cultivars over green ash trees (Dirr 2009, author's personal communication with green industry professionals). The more desirable growth habit, urban tolerance, and brilliant fall color of white ash, and drought tolerance and dark green leaves of blue ash are probably important factors in their preference and decision-making (Schlesinger 1990, Griffith 1991, Dirr 2009).

**Efficacy of dinotefuran, clothianidin, and dinotefuran combined with clothianidin for protection of ash trees from EAB.** In the single Glen Ellyn study, dinotefuran and clothianidin used alone, and in combination, were equally and very effective in protecting ash trees up to 50 cm dbh (PCL ratings less than 18%). There does not appear to be any significant additive effect by combining dinotefuran with clothianidin. These same active ingredients were as effective as imidacloprid used alone or when imidacloprid is combined with clothianidin for protecting similar size ash trees (PCL ratings of 10% to 14%).

Interestingly, the treated Glen Ellyn green ash trees did not show any significant effects from the 2012 drought. It should be noted, however, that the trees at the Glen Ellyn site had been treated for two years prior to the 2012 drought. Further, dinotefuran (Safari) is much more water soluble than imidacloprid and is able to be taken up and distributed faster in the tree compared with imidacloprid, allowing for dinotefuran to quickly target EAB larval feeding particularly when applied later in the growing season (Tattar et al. 1998; USEPA. U.S. Environmental Protection Agency 2004, 2014; Extoxnet Extension Toxicology Network 2010; Bryne et al. 2012, 2014; Nix et al. 2013; Bonmatin et al. 2015; Mach et al. 2018). This provides for greater flexibility for treating trees once an infestation has been identified, even during drought conditions. However, one disadvantage is that dinotefuran has a shorter residual efficacy compared to imidacloprid or emamectin benzoate.

Scheduling of applications within a given growing season is also important. In our studies, the single July basal soil drench of dinotefuran was equally effective in protecting trees as the June application, and is consistent with previous studies by McCullough et al. (2011, 2019) and Smitley et al. (2015). In situations where an EAB infestation is not discovered until later in the growing season (i.e. mid-June to mid-July), and/or if treatments are otherwise delayed, dinotefuran or clothianidin alone, and/or in combination can be an effective option for protecting ash trees up to 50 cm dbh.

**Effectiveness of emamectin benzoate for protection of ash trees from EAB.** After eight years, it is evident that all rates, application timing, and trunk injection application methods of professional Tree-Age (emamectin benzoate) is highly effective (PCL less than 6%) in protecting green ash trees up to 55 cm dbh, even during a severe drought. Our findings are comparable to

studies conducted by Anulewicz et al. (2007, 2008), Bick et al. (2018), Lewis and Turcotte (2015), McCullough et al. (2011, 2019), and Smitley et al. (2010 a, b). Additionally, in an unrelated ash Naperville–Bolingbrook, Illinois tree preservation study, biennial trunk injection applications of Tree-Age (emamectin benzoate) is currently providing excellent protection of similar size green and white ash trees (Rutkowski and Mitz 2017, Miller and Gould 2018).

**In summary**, while some ash trees have recovered from as much as 60% canopy loss (Smitley et al. 2007), members of the green industry generally consider up to 30% canopy loss to be acceptable level of damage when evaluating for treatment efficacy and whether to remove and replace a tree (Bick et al. 2018). The vast majority of the treatments evaluated in this study provided good to excellent control (percent canopy loss less than 30%), with the exception of BATSGF1and BATSC2X at the Aurora site, and BATSCF at the Hinsdale site. Annual applications of imidacloprid applied alone at 0.57 g active ingredient (a.i.)/2.54 cm dbh or greater, clothianidin and dinotefuran applied alone at 0.93 g a.i./2.54 cm dbh or greater, imidacloprid + clothianidin at 0.57 g a.i. + 0.28 g a.i.2.54 cm dbh or greater, or dinotefuran + clothiandin applied annually at 0.47 g a.i. + 0.46 g a.i./2.54 cm dbh or greater provided good to excellent protection of green, white, and blue ash trees up to 61cm dbh. Biennial trunk injections of emamectin benzoate applied at 0.2 to 0.6 g a.i./2.54 cm dbh provided good to excellent protection of green ash trees with a mean dbh of 61 cm and, may provide extended protection under moderate to heavy EAB pressure even during a record-setting drought. Tree care practitioners and homeowners have a variety of available options for the timing and application of protective chemicals including basal soil drenches, basal broadcast applications, basal trunk sprays, and soil and trunk injections for protection of their ash tree resource from the EAB. It is important to remember that not all ash tree species are equally susceptible to EAB, and studies are showing, that while still susceptible, decline and death in white and blue ash is slower, and in some cases they may actually survive an EAB infestation (Tanis and McCullough 2012, Robinett and McCullough 2019). Further, professionals and homeowners should recognize the impact that abiotic factors (i.e. drought), and soil rooting volumes may have on the uptake and distribution of protective systemic chemicals, specifically where EAB infestations have been initially confirmed. Diligent inspection of susceptible ash trees, proper selection, timing and application of insecticidal treatments, public awareness and education, and communication among all concerned parties are all critical to implementing an effective EAB management plan for protection of our valuable urban forest resource.

## Acknowledgments

The authors would like to express their sincere thanks to E. Zimmerman, Village of Skokie, J. Trousethick and B. Doerr, Village of Homewood, W. Pauley, City of Aurora, M. Collins, Village of Riverside, J. Finnel, Village of Hinsdale, J. Mitz, City of Naperville, B. Santore, Village of Woodridge, P. Drescher, Village of Glen Ellyn, and R. Walton and R. Lootens, Fermi National Accelerator Laboratory, Batavia, Illinois for their assistance and cooperation in allowing us to use their trees for study, and for providing staff and equipment. We would like to sincerely thank J. Doccola and J. Aiken of ArborJet Inc., S. Darnell of Bayer Corporation, S. Bernick of Rainbow Treecare Scientific Advancements, and J. Chamberlain and J. Fausey of Valent Corporation for assisting with applications, and providing funding and material in support of these studies. Thanks are due to N. Dahlberg, S. Kolb, L. Ivanyi, D. Kusnierz, and other members of The Morton Arboretum entomology lab for the assistance in applying treatments and tree canopy assessments. A special note of thanks is extended to B. Overbey for her very able assistance with data entry, editing, and formatting of the manuscript.

## Literature Cited

**Anulewicz, A.C., D.G. McCullough, and D.L. Cappaert. 2007**. Emerald ash borer (*Agrilus planipennis*) density and canopy dieback in three North American ash species. Arboriculture and Urban Forestry 33: 338–349.

**Anulewicz, A. C., D.G. McCullough, D.L. Cappaert, and T.M. Poland. 2008**. Host range of the emerald ash borer (*Agrilus planipennis* Fairmaire) (Coleoptera: Buprestidae) in North America: Results of multiple-choice field experiments. Environmental Entomology 37(1): 230–241.

**Ball, J., and G. Simmons. 1980**. The relationship between bronze birch borer and birch dieback. Journal of Arboriculture 6: 309–314.

**Bauer, L.S., J.J. Duan, J.R. Gould, and R. Van Driesche. 2015**. Progress in the classical biological control of *Agrilus planipennis* Fairmaire (Coleoptera: Buprestidae) in North America. Canadian Entomologist 147: 300–317.

**Bick, E.N., N.J. Forbes, C. Haugen, G. Jones, S. Bernick, and F. Miller. 2018.** Seven-year evaluation of insecticide tools for emerald ash

borer in *Fraxinus pennsylvanica* (Lamiales: Oleaceae) trees. Journal of Economic Entomology 111 (2): 732–740.

**Bonmatin, J.M., C. Giorio, V. Giolami, D. Goulson, D. Kreutzweiser, D.P., C. Krupke, M. Liess, E. Long, M. Marzaro, A.D. Mitchell, D.A. Noome, N. Simon-Delso, and A. Tapparo. 2015.** Environmental fate and exposure: neonicotinoids and fipronil. Environmental Science and Pollution Research 22: 35–67.

**Byrne, F.J., A. Urena, L.J. Robinson, R.I. Krieger, J. Doccola, J.G. Morse. 2012**. Evaluation of neonicotinoid, organophosphate, avermectin trunk injections for management of avocado thrips in California avocado groves. Pest Management Science 68: 811–817.

**Byrne, F. J., Krieger, J. Doccola, and J.G. Morse. 2014**. Seasonal timing of neonicotinoid and organophosphate trunk injections to optimize the management of avocado thrips in California avocado groves. Crop Protection 57: 20–26.

**Cappaert, D. L., D. G. McCullough, T. M. Poland, and N.W. Siegert. 2005**. Emerald ash borer in North America: A research and regulatory challenge. American Entomologist 51: 152–165.

**Chen, Y.T., and T.M. Poland. 2009.** Biotic and abiotic factors affect green ash volatile production and emerald ash borer adult feeding preference. Environmental Entomology 38(6): 1756–1764.

**Chen, Y.T., and T.M. Poland. 2010.** Nutritional and defensive chemistry of three North American ash species: Possible roles in performance by ash borer adults. The Great Lakes Entomologist 43: 20–33.

**Chen, Y. T. Ciaramitaro, and T.M. Poland. 2011**. Moisture content and nutrition as selection forces for emerald ash borer larval feeding behavior. Ecological Entomology 36: 344–354.

**Chen, Y., M.D. Ulyshen, and T.M. Poland. 2012.** Differential utilization of ash phloem by emerald ash borer larvae: Ash species and larval stage effects. Agricultural and Forest Entomology 14: 324–330.

**Cregg, B.M., and M.E. Dix. 2001**. Tree moisture stress and insect damage in urban areas in relation to heat island effects. Journal of Arboriculture 27: 8–17.

**Creticos, P.A. 2013**. An economic impact analysis of alternative municipal responses to an emerald ash borer infestation. Institute for work and the economy. Regional Development Institute. Northern Illinois University, DeKalb, Illinois. 7 pp.

**Craul, P. 1999.** Urban soils: application and practices. New York, NY. John Wiley and Sons. 377 pp.

**Dirr, M.A. 2009.** Manual of woody landscape plants: Their identification, ornamental characteristics, culture, propagation, and uses. Stipes Publishing Company, Champaign, Illinois. 826 pp.

**Duan, J.J., T.J. Watt, and K. Larson. 2014**. Biology, life history, and laboratory rearing of *Spathius galinae* (Hymenoptera: Braconidae), a larval parasitoid of the invasive emerald ash borer (Coleoptera: Buprestidae). Journal of Economic Entomology 107(3): 939–946.

**Eyles, A., W. Jones, K. Riedl, D. Cipollini, S. Schwartz, K. Chan, D.A. Herms, and P. Bonello. 2007**. Comparative phloem chemistry of Manchurian (*Fraxinus mandshurica*) and two North American ash species (*Fraxinus americana* and *F. pennsylvanica*). Journal of Chemical Ecology 33: 1430–1448.

**Extoxnet, Extension Toxicology Network. 2010.** Pesticide information profile. Imidacloprid. (http://pmep.cce.cornell.edu/profiles/extoxnet/halosyfop-methylparathion/imidacloprid-ext.html).

**Fahey, R.T., M.B. Bialecki, and D.R. Carter. 2013.** Tree growth and resilience to extreme drought across an urban land-use gradient. Arboriculture and Urban Forestry 39(6): 279–285.

**Flower, C.E., J.E. Dalton, K.S. Knight, M. Brikha, and M.A. Gonzalez-Meler. 2015**. To treat or not to treat: Diminishing effectiveness of emamectin benzoate injections in ash trees heavily infested by emerald ash borer. Urban Forestry and Urban Greening 14: 790–795.

**Gregory, J., and M. Dukes. 2006.** Effect of urban soil compaction on infiltration rate. Journal of Soil and Water Conservation 61: 117–124.

**Griffith, R.S. 1991.** *Fraxinus americana*. In: W.C. Fischer. USDA Forest Service, Intermountain Research Station. Missoula, Montana.

**Haack, R. A., E. Jendek, H. Liu, K. R. Marchant, T. R. Petrice, T. M. Poland and H. Ye. 2002.** The emerald ash borer: A new exotic pest in North America. Newsletter of the Michigan Entomological Society 47(3/4): 1–5.

**Hauer, R.J., and W.D. Peterson. 2017.** Effects of emerald ash borer on municipal forestry budgets. Landscape Urban Planning 157: 98–105.

**Herms, D.A. 2002.** Strategies for deployment of insect resistant ornamental plants. In: M.R. Wanger, K.M. Clancy, F. Lieutier, and T.D. Paine (Eds.). *Mechanisms and deployment of resistance in trees to insects*. Pp. 217–237.

Kluwer Academic Press, Dordrecht, The Netherlands. 332 pp.

**Herms, D.A. 2015.** Host range and host resistance. Pp. 153–163. In: R.G. Van Driesche, and R. Reardon (Eds.). Biology and control of emerald ash borer. Technical Bulletin FHTET 2014-09, USDA Forest Service, Morgantown, West Virginia, USA. <www.fs.fed.us/foresthealth/technology/pdfs/FHTET-2014-09_Biology_Control_EAB.pdf>

**Herms, D. A., and D. G. McCullough. 2014.** Emerald ash borer invasion of North America: History, biology, ecology, impacts, and management. Annual Review of Entomology 59: 13–30.

**Herms D.A., D.G. McCullough, D.R. Smitley, C. Sadof, R.C. Williamson, and P.L. Nixon. 2009**. Insecticide options for protecting ash trees from emerald ash borer. North Central IPM Center Bulletin. 1st Edition. 12 pp.

**Herms D.A., D.G. McCullough, D.R. Smitley, C. Sadof, R.C. Williamson, and P.L. Nixon. 2014**. Insecticide options for protecting ash trees from emerald ash borer. North Central IPM Center Bulletin. 2nd Edition. 12 pp.

**Herms D.A., D.G. McCullough, C.S. Sadof, D.R. Smitley. F.D. Miller, and C.W. Cranshaw. 2019**. Insecticide options for protecting ash trees from emerald ash borer. North Central IPM Center Bulletin. 3rd Edition. 16 pp.

**Huberty, A.F., and R.F. Denno. 2004.** Plant water stress and its consequences for herbivorous insects: A new synthesis. Ecology 85: 1383–1398.

**Jandel Scientific. 1992**. SigmaStat for Windows. San Rafael, CA. 232 pp.

**Kovacs, K.F., R.G. Haight, D.G. McCullough, R.J. Mercader, N.W. Siegert, and A.M. Liebhold. 2010.** Cost of potential emerald ash borer damage in U.S. communities, 2009-2019. Ecological Economics 69: 569–578.

**Larsson, S. 1989.** Stressful times for the plant stress-insect performance hypothesis. Oikos 65: 277–283.

**LeGoff, N., and J.M. Ottorini. 1996.** Leaf development and stem growth in ash (*Fraxinus excelsior*) as affected by tree competitive status. Journal of Applied Ecology 4: 793–802.

**Lewis, P.A., and R.M. Turcotte. 2015.** Use of chemical protection and host tree reduction to control an emerald ash borer infestation in West Virginia. Journal of Biodiversity Management and Forestry 4:1. Available from http://dx.doi.org/10.4172/2327-4417.1000136

**Loerch, C.R., and E.A. Cameron. 1984**. Within-tree distribution and seasonality of immature stages of the bronze birch borer, *Agrilus anxius* (Coleoptera: Buprestidae). Canadian Entomologist 116: 147–152.

**Mach, B.M., S. Bondarenko, and D.A. Potter. 2018**. Uptake and dissipation of neonicotinoid residues in nectar and foliage of systemically treated woody landscape plants. Environmental Toxicology and Chemistry 37(3): 860–870.

**Marshall, J.M., E.L. Smith, R. Mech, A.J. Strober. 2013**. Estimates of *Agrilus planipennis* infestation rates and potential survival of ash. American Midland Naturalist 169: 179–193.

**Martinson, H., C.S. Sargent, and M.J. Raupp. 2014**. Tree water stress and insect geographic origin influence patterns of herbivory in green (*Fraxinus pennsylvanica*) and Manchurian (*F. mandshurica*) ash. Arboriculture and Urban Forestry 40 (6): 322–344.

**McCullough, D.G., and R.J. Mercader. 2012**. Evaluation of potential strategies to Slow Ash Mortality (SLAM) caused by emerald ash borer (*Agrilus planipennis*): SLAM in an urban forest. International Journal of Pest Management 58(1): 9–23.

**McCullough, D.G., and N.W. Siegert. 2007**. Estimating potential emerald ash borer (Coleoptera: Buprestidae) populations using ash inventory data. Journal of Economic Entomology 100: 1577–1586.

**McCullough, D. G., T. M. Poland, A. C. Anulewicz, and D. L. Cappaert. 2011**. Evaluation of *Agrilus planipennis* (Coleoptera: Buprestidae) control provided by emamectin benzoate and two neonicotinoid insecticides, one and two seasons after treatment. Journal of Economic Entomology 104: 1599–1612.

**McCullough, D.G., T.M. Poland, A.R Tluczek, A. Anulewicz, J. Wieferich, and N.W. Siegert. 2019.** Emerald ash borer (Coleoptera: Buprestidae) densities over a 6-yr period on untreated trees and trees treated with systemic insecticides at 1-, 2-, and 3-yr intervals in a central Michigan forest. Journal of Economic Entomology 112(1): 201–212.

**McKenney, D.W., J.H. Pedlar, D. Yemshanov, D. Barry Lyons, K.L. Campbell, and K. Lawrence. 2012.** Estimates of the potential cost of emerald ash borer (*Agrilus planipennis* Fairmaire) in Canadian municipalities. Arboriculture and Urban Forestry 38: 81–91.

**Miller, F., and J. Gould. 2018**. Compatibility of insecticides and biocontrol for controlling EAB in urban environments. USDA-APHIS Grant Project Progress Report. 2 pp.

**Nielsen, D.G., V.L. Muilenburg, and D.A. Herms. 2011.** Interspecific variation in resistance of Asian, European, and North American birches (*Betula* spp.) to bronze birch borer (Coleoptera: Buprestidae). Environmental Entomology 40(3): 648–653.

**Nix K., P. Lamdin, J. Grant, C. Coots, and P. Merten. 2013**. Concentration of levels of

imidacloprid and dinotefuran in five tissues types of black walnut, *Juglans nigra*. Forests 4: 811–817.

**Poland, T.M., and D.G. McCullough. 2006.** Emerald ash borer: invasion of the urban forest and the threat to North American ash resources. Journal of Forestry 104: 118–124.

**Poland, T.M., Y. Chen, J. Kock, and D. Pureswaran. 2015.** Review of the emerald ash borer (Coleoptera: Buprestidae), life history, mating behaviours, host plant selection, and host resistance. The Canadian Entomologist 147: 252–262.

**Raupp, M. J., A. B. Cumming, and E. C. Raupp. 2006**. Street tree diversity in eastern North American and its potential for tree loss to exotic borers. Arboriculture and Urban Forestry 32: 297–304.

**Rebek, E.J., D.A. Herms, and D.R. Smitley. 2008.** Interspecific variation in resistance to emerald ash borer (Coleoptera: Buprestidae) among North American and Asian ash (*Fraxinus* spp.). Environmental Entomology 37: 242–246.

**Robinett, M.A., and D.G. McCullough. 2019**. White ash (*Fraxinus americana*) survival in the core of the emerald ash borer (*Agrilus planipennis*) invasion. Canadian Journal of Forest Research 49(5): 510–520. Available from https://doi.org/10.1139/cjfr-2018-0320

**Rutkowski, J., and J. Mitz. 2017**. Emerald ash borer program. Manager's memorandum: Council work request report, City of Naperville, Illinois. 4 pp. Available from https://www.naperville.il.us/residents/our-urban-forest/emerald-ash-borer

**Sadof, C. S., L. Purcell, F. J. Bishop, C. Quesada, and Z. Zhang. 2011**. Evaluating restoration capacity and costs of managing the emerald ash borer with a web-based cost calculator in urban forests. Arboriculture and Urban Forestry 37: 74–83.

**Schlesinger, R.C. 1990**. *Fraxinus americana* L. White ash. Pp. 333-338 in R.M. Burns and B.H. Honkala. Silvics of North America. Volume 2. Hardwoods. USDA Forest Service Agriculture Handbook 654. Washington, D.C.

**Showalter, D.N., C. Villari, D.A. Herms, and P. Bonello. 2018.** Drought stress increased survival and development of emerald ash borer larvae on coevolved Manchurian ash and implicates phloem-based traits in resistance. Agricultural and Forest Entomology 20: 170–179.

**Smitley, D.R., T. Davis, K. Newhouse, and E. Rebek. 2007.** Evaluation of insecticide products for control of emerald ash borer, p. 47, *In* V. Mastro, D. Lance, R. Reardon, and G. Parra (eds.), Proceedings of the Emerald Ash Borer and Longhorned Beetle Research and Technology Development Meeting, 23=24 October, 2007, Pittsburgh, PA., USDA Forest Service Forest Health Technology Enterprise Team FHTET: 2007-04, Morgantown, WV.

**Smitley, D. R., T. W. Davis, and E. J. Rebek. 2008**. Progression of ash canopy loss outward from the initial infestation of emerald ash borer (Coleoptera: Buprestidae) in Southeast Michigan. Journal of Economic Entomology 101: 1643–1650.

**Smitley, D. R., J. J. Doccola, and D. L. Cox. 2010a**. Multiple year protection of ash trees from emerald ash borer, *Agrilus planipennis* Fairmaire (Coleoptera: Buprestidae), with a single trunk injection of emamectin benzoate and single-year protection with an imidacloprid basal drench. Arboriculture and Urban Forestry 36: 206–211.

**Smitley, D. R., E. J. Rebek, R. N. Royalty, T. W. Davis, and K. F. Newhouse. 2010b**. Protection of individual ash trees from emerald ash borer (Coleoptera: Buprestidae) with basal soil applications of imidacloprid. Journal of Economic Entomology 103: 119–126.

**Smitley, D.R., D.A. Herms, and T.W. Davis. 2015**. Efficacy of soil applied neonicotinoid insecticides for long-term protection against emerald ash borer (Coleoptera: Buprestidae). Journal of Economic Entomology 108(5): 2344–2353.

**Subburayalu, S.K., and D. Syndor. 2018**. Results and implications following a twelve-year monitoring of ash (*Fraxinus* spp.) mortality due to *Agrilus planipennis* in an urban forest plantation. Arboriculture and Urban Forestry 44(3): 154–160.

**Sydnor, T.D., M. Bumgardner, and A. Todd. 2007**. The potential economic impacts of emerald ash borer (*Agrilus planipennis*) on Ohio communities. Arboriculture and Urban Forestry 33(1): 48–54.

**Tanis, S.R., and D.G. McCullough. 2012.** Differential persistence of blue ash and white ash following emerald ash borer invasion. Canadian Journal of Forest Research 42(8): 1542–1550.

**Tanis, S.R., and D.G. McCullough. 2015**. Host resistance of five *Fraxinus* species to *Agrilus planipennis* (Coleoptera: Buprestidae) and effects of paclobutrazol and fertilization. Environmental Entomology 44(2): 287–299.

**Tattar, T.A., J.A. Dotson, M.S. Ruizzo, and V.B. Steward. 1998**. Translocation of imidacloprid in three tree species when trunk and soil-injected. Journal of Arboriculture 24(1): 54–56.

**USEPA. U.S. Environmental Protection Agency. 2004**. Pesticide fact sheet. Dinotefuran. U.S. Environmental Protection Agency. Washington, D.C.

**USEPA. US Environmental Protection Agency. 2014**. Guidance for assessing pesticide risks to bees. [cited 2017 Aug 1]. Available from: https://www.epa.gov/sites/production/files/2014-06/documents/pollinator_risk_assessment_guidance_06_19_14.pdf.

**Vannatta, A.R., R. H. Hauer, and N.M. Schuettpelz. 2012.** Economic analysis of emerald ash borer (Coleoptera: Buprestidae) management options. Journal of Economic Entomology 105: 196–206.

**Wang, B., R. Gao, V. C. Mastro, and R. C. Reardon. 2005**. Toxicity of four systemic neonicotinoids to adults of *Anopophora glabripennis* (Coleoptera: Cerambycidae). Journal of Economic Entomology 98: 2292–2300.

**Wang, X.Y., L.M. Cao, Z.Q. Yang, J.J. Duan, J.R. Gould, and L.S. Bauer. 2016**. Natural enemies of emerald ash borer (Coleoptera: Buprestidae) in northeast China with notes on two species of parasitic Coleoptera. Canadian Entomologist 148(3): 329–342.

# Reinstatement of *Andrena vernalis* Mitchell (Hymenoptera: Andrenidae) from Synonymy with *A. ziziae* Robertson

Zachary M. Portman[1,*], Ian G. Lane[1], Gabriella L. Pardee[1], Daniel P. Cariveau[1]

[1] Department of Entomology, University of Minnesota, 1980 Folwell Ave., St. Paul, MN 55108 USA.
[*] Corresponding author: (e-mail: zportman@umn.edu)

**Abstract**

*Andrena* (*Micrandrena*) *ziziae* Robertson, 1891 (Hymenoptera: Andrenidae) is a well-known species found in a variety of habitats in the eastern and central United States and adjacent southern Canada. *Andrena* (*Micrandrena*) *vernalis* Mitchell, 1960 was described from five female specimens in the eastern United States and was synonymized with *A. ziziae* by Ribble in 1968. Recently collected specimens from throughout Minnesota have revealed that *A. ziziae sensu* Ribble is actually two species, one of which matches *A. vernalis*. Here, we reinstate *A. vernalis* as a valid species and describe the previously unknown male. We provide diagnostic characters that separate *A. ziziae* and *A. vernalis*, as well as data on the geographic range and floral preferences of both species in Minnesota. *Andrena vernalis* appears to be restricted to high-quality remnant habitats, making it a species of potential conservation concern. These changes will require that previous work on *A. ziziae* be revisited to determine if *A. vernalis* is also present.

**Key words:** Species complex, specialization, *Zizia*, Apiaceae, taxonomy

---

*Andrena* (*Micrandrena*) *ziziae* Robertson, 1891 (Hymenoptera: Andrenidae) is a small, solitary, ground nesting bee (Ribble 1968). *Andrena ziziae* collects pollen only from plants in the family Apiaceae (Ribble 1968, Wood and Roberts 2018), making it an important pollinator of forb species in the genus *Zizia* and *Thaspium* (Lindsey 1984, Lindsey and Bell 1985). *Andrena ziziae* occurs throughout the eastern and central United States and adjacent southern Canada (Ribble 1968). It can be locally abundant where individuals of its host plants persist, and is found in a variety of habitats, including urban areas (Normandin et al. 2017), tallgrass prairie (Davis et al. 2008), prairie restorations (Harmon-Threatt and Hendrix 2015), powerline strips (Russell et al. 2005), and montane and piedmont areas (Lindsey 1984).

*Andrena ziziae* was originally described by Robertson (1891). Soon after, Robertson (1897) split *A. ziziae* into two species, describing the second species as *A. personata* Robertson, 1897. *Andrena* (*Micrandrena*) *vernalis* Mitchell, 1960 was described from five female specimens (the males were unknown) from Connecticut (type locality), Ohio, and New York. The characters Mitchell (1960) used to separate females of *A. vernalis* from *A. ziziae* (as well as the closely related *Micrandrena* Ashmead species *A. illinoiensis* Robertson, 1891 and *A. salictaria* Robertson, 1905) were the broader facial fovea and the presence of a unique pleural ridge, which he described as "pleura protuberant below, having a flattened triangular area anterior to mid coxal cavities, delimited by a distinct ridge".

*Andrena vernalis* was later synonymized with *A. ziziae* by Ribble (1968) as part of a broader revision of the subgenus *Micrandrena*. Ribble (1968) justified the synonymy by stating that there was continuous variation in the structure of the pleural ridge and specimens both with and without the pleural ridge co-occurred throughout the range of *A. ziziae*. However, Ribble (1968) did not examine the holotype of *A. vernalis* before synonymizing the species (though it appears he did examine two paratypes). In addition, the illustrations of the male terminalia of *A. ziziae* in Ribble (1968) differ substantially from the illustrations of Mitchell (1960).

Recent collections throughout Minnesota provided hundreds of specimens of *A. ziziae*, including many specimens that match Mitchell's (1960) description of *A. vernalis*. Examination of these specimens revealed that in addition to the pleural ridge, female specimens matching *A. vernalis* have multiple other consistent morphological characters that separate them from the typical form of *A. ziziae*, indicating that *A. vernalis* is a valid species in its own right. This conclusion is reinforced by the discovery of the male of *A. vernalis*, including both

novel specimens and a male collected with the type series. The males of *A. vernalis* have clear and consistent differences in the male terminalia that further cement its status as a valid species.

Here, we resurrect *A. vernalis* from synonymy and provide additional diagnostic characters that separate it from *A. ziziae*. In addition, we provide the first description of the previously unknown male. Although *A. vernalis* appears to occur across the eastern United States, we focus here on Minnesota, where it is important to provide a clear species definition for upcoming ecological studies. Our data suggest that both *A. ziziae* and *A. vernalis* specialize on *Zizia* pollen in Minnesota prairies. However, unlike *A. ziziae*, *A. vernalis* appears to be associated with high-quality remnant habitats, making it essential to further delineate the range and habitat requirements of these two species.

## Methods & Materials

The following abbreviations are used for museums and institutions:

| | |
|---|---|
| INHS | Illinois Natural History Survey, Champaign, IL, USA. Dr. Thomas McElrath. |
| MCZ | Museum of Comparative Zoology, Harvard University, Cambridge, MA, USA. Dr. Crystal Maier. |
| MNDNR | Minnesota Department of Natural Resources, St. Paul, MN, USA. Dr. Jessica Petersen. |
| NCSU | North Carolina State University Insect Museum, Raleigh, NC, USA. Dr. Bob Blinn. |
| UMSP | University of Minnesota Insect Collection, St. Paul, MN, USA. Dr. Robin Thomson. |

We examined 850 specimens for this study. Specimens were largely drawn from a mix of published (Pennarola 2019) and unpublished ecological studies of the Minnesota bee fauna. The type of *A. vernalis* was examined at MCZ and additional non-type specimens were examined from the type locality. Unless otherwise noted, specimens are deposited in the UMSP or the Cariveau native bee lab collection at the University of Minnesota and will later be deposited at UMSP.

Classifications and terminology follow Michener (2007), except wing vein terminology follows Engel (2001). The abbreviations S1…S8 and T1…T7 are used for sterna and terga, respectively. The format and terminology of specimen descriptions are modified and adapted from Mitchell (1960) and Ribble (1968). Specimen images and measurements were taken with an Olympus DP27 camera mounted on an Olympus SZX16 stereomicroscope. Images were stacked with CombineZP software (Hadley 2010). Photographic plates were compiled using Adobe Photoshop 2018 software (Adobe Systems Inc., San Jose, CA). Maps were created using ArcMap software v10.5.1 (ESRI, Redlands, CA).

*Pollen analysis.* To gain insight into foraging preferences for *A. ziziae* and *A. vernalis*, we used hand nets to collect bees that were actively foraging on *Zizia aptera* and *Z. aurea* from eight prairies throughout western MN. We placed all collected bees into individual 1.5 mL microcentrifuge tubes and cleaned the net before continuing our sampling to reduce pollen contamination. All collected specimens were sacrificed in the freezer, then swabbed for pollen, and later identified to species.

In the lab, we sampled the pollen on each specimen by swabbing the head, thorax, abdomen, and underside of the thorax with a small piece (~8 $mm^3$) of fuchsin gel (Kearns and Inouye 1993). Next, we melted the fuchsin gel onto a microscope slide and covered it with a cover slip. We then used a compound microscope to count the first 300 pollen grains encountered on the slide (Ritchie et al. 2016), recording the number of *Zizia* pollen grains and the number of pollen grains from other genera. We then calculated the mean number of *Zizia* pollen grains and the mean proportion of *Zizia* pollen grains bees carried on their body for each species. We were not able to differentiate between the pollen of *Z. aurea* and *Z. aptera*.

## Results

### *Andrena* (*Micrandrena*) *vernalis* Mitchell, new status

Fig. 1A,C,E, 2A,C,E, 3A,C,E, 4A,C,E, 5A–B, 6A–C, 7, 8A,C, 9

*Andrena vernalis* Mitchell 1960: 168, ♀. Holotype ♀, USA, CT, Colebrook, 31 May 1922, W.M. Wheeler leg. (MCZ 30485).

*Andrena ziziae* (in part): Ribble, 1968: 267 (syn. *A. vernalis* with *A. ziziae*).

**Diagnosis.** Female *A. vernalis* can most easily be separated from *A. ziziae* by the broader facial fovea (Fig. 1A–D) and the more prominent tergal hair bands (Fig. 1E–F). The fovea of *A. vernalis* take up nearly the entire distance between the eye and lateral ocelli (Fig. 1C), whereas the fovea of *A. ziziae* take up only slightly more than

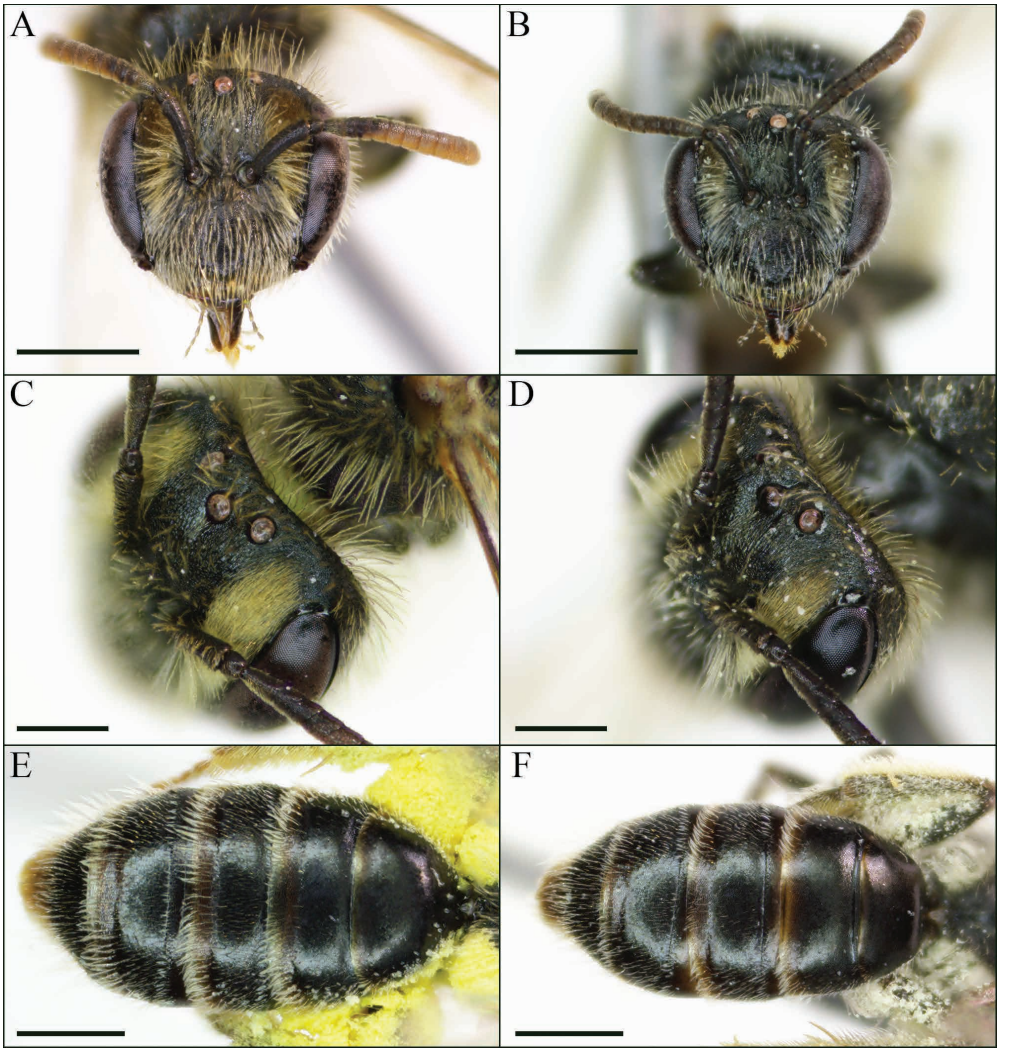


Figure 1. *Andrena vernalis* (left column) and *A. ziziae* (right column) female face and abdominal characters: (A) *A. vernalis* face; (B) *A. ziziae* face; (C) *A. vernalis* facial fovea; (D) *A. ziziae* facial fovea; (E) *A. vernalis* abdomen; (F) *A. ziziae* abdomen. Scale bars: A–B, E–F = 1mm; C–D = 500 µm.

half the distance between the eye and lateral ocelli (Fig. 1D). The apical tergal hair bands of *A. vernalis* are longer (Fig. 1E) than the relatively close-cropped hairs of *A. ziziae* (Fig. 1F), though these hairs can be worn off. In addition, tergal rims of *A. vernalis* are distinctly transparent on nearly the entire apical impressed area (Fig. 1E, 2A), whereas *A. ziziae* have the tergal rims are transparent only at the very apex (Fig. 1F).

Other characters that can help separate female *A. vernalis* from *A. ziziae* include the propodeum of *A. vernalis*, which has a slightly more well-defined triangle with the interior of the triangle rugulose (Fig. 2E). In contrast, the triangle of *A. ziziae* is less well-defined and less strongly sculptured (Fig. 2F). In addition, the Rs vein of *A. vernalis* is generally attached to the marginal cell 2–3 vein widths from the stigma (Fig. 3A) whereas the Rs vein of *A. ziziae* is generally attached about 1 vein width from the stigma (Fig. 3B). However, this character is variable and some *A. ziziae* have the Rs vein attached slightly further, about 2 vein widths (Fig. 3D), overlapping with some

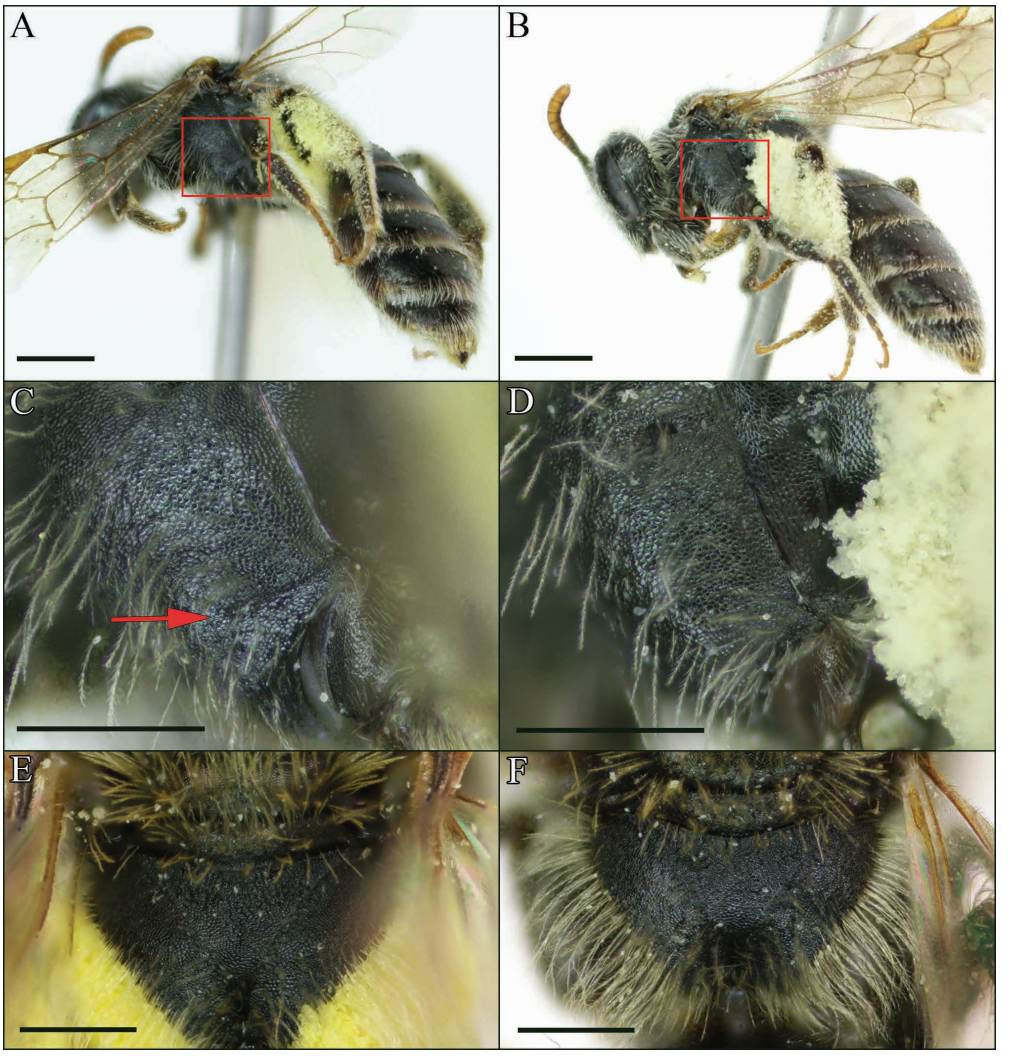


Figure 2. *Andrena vernalis* (left column) and *A. ziziae* (right column) female thoracic characters: (A) *A. vernalis* lateral view; (B) *A. ziziae* lateral view; (C) *A. vernalis* closeup of pleural depression and ridge, indicated by red arrow; (D) *A. ziziae* closeup of smoothly rounded pleura; (E) *A. vernalis* propodeal triangle; (F) *A. ziziae* propodeal triangle. Scale bars: A–B = 1mm; C–E = 500 µm.

*A. vernalis* that have the Rs vein attached closer to the stigma (Fig. 3C).

Finally, in *A. vernalis*, the pleura directly anterior to the mid-coxa has a depressed subtriangular area delineated by a weakly carinate but distinct ridge (Fig. 2A,C, 9A–B). In contrast, *A. ziziae* has the pleura smoothly rounded (Fig. 2B,D), or at most weakly protruding in this area. While this character is diagnostic, it can be extremely difficult to see, especially since it is typically blocked from view by the midlegs.

Females of *A. vernalis* key out correctly in Mitchell (1960), and they key out to *A. ziziae* in Ribble (1968) and can be separated by the previously mentioned characters.

Male *A. vernalis* can be separated from *A. ziziae* by their broader antennal segments (measured on the anterior side as in Fig. 4B): F3–7 of *A. vernalis* are distinctly broader than long (Fig. 4A,C), whereas in *A. ziziae*, F3–7 are longer than broad or have length and width equal (Fig. 4B,D). In addition, the apical lobes of S7 are truncate in *A. vernalis* (Fig. 5B) and rounded in *A. ziziae* (Fig. 5D),

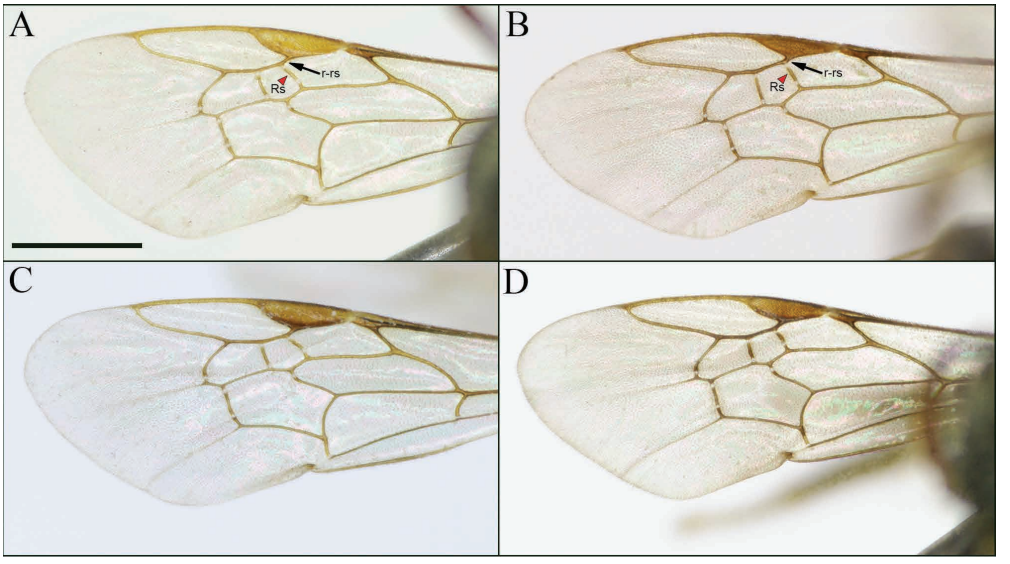


Figure 3. *Andrena vernalis* (left column) and *A. ziziae* (right column) forewings: (A) typical *A. vernalis* female forewing with longer r-rs crossvein (long black arrow) and Rs vein (short red arrow) attached to marginal cell 2–3 vein widths from stigma; (B) typical *A. ziziae* female forewing with shorter r-rs crossvein (long black arrow) and Rs vein (short red arrow) attached to marginal cell about 1 vein width from stigma; (C) *A. vernalis* female forewing with Rs vein attached more closely to stigma, about 2 vein widths; (D) *A. ziziae* female forewing with Rs vein attached further from stigma, about 2 vein widths. Scale bar = 1 mm, all images are at the same scale.

and the dorsal lobes of genitalia are much broader in *A. vernalis* (Fig. 6A) than *A. ziziae* (Fig. 6D).

Additional characters can help separate males of *A. vernalis* and *A. ziziae* but they are subtle and often difficult to discern. Similar to females, male *A. vernalis* have a slightly more well-defined propodeal triangle, the apical rims of the terga are more broadly transparent in *A. vernalis* (Fig. 4E) than *A. ziziae* (Fig. 4F), the Rs wing vein attaches further from the stigma in *A. vernalis* (as in Fig. 3A) than *A. ziziae* (as in Fig. 3B) though this character can be variable, and the pleura directly anterior to the mid-coxa has a depressed subtriangular area delineated by a slightly protruding margin (Fig. 9C–D). Males are difficult to separate from *A. ziziae* and it is recommended that the terminalia be used to confidently identify them.

Males of *A. vernalis* key out to *A. ziziae* in both Mitchell (1960) and Ribble (1968) and can be separated by the previously mentioned characters.

**Description of male.** Body length: 6.0 mm, range 5.8–6.2 mm; ITD: 1.25 mm (n=7).

*Integumental color.* Body black without any metallic reflections; clypeus yellow except for two small, sublateral black spots and dark apical rim (Fig. 4A); antennae dark brown above and light brown below (Fig. 4C); wing veins a mix of light and dark brown (Fig. 3E); tibial spurs clearish-white; apical tarsi light brown; basitarsi generally with apex somewhat light brown; apical rims of terga hyaline (Fig. 4E).

*Structure.* Length and breadth of facial quadrangle about equal; eyes slightly converging below (Fig. 4A); clypeus very slightly convex, projecting slightly below lower margin of eye, shining with faint tessellation, punctures small and obscure, separated by about 2–3 puncture widths, midline of clypeus more sparsely punctate; vertex less than one ocellar diameter; cheeks subequal to eyes in width, rounded posteriorly; malar space linear; F1 subequal to F2+F3; middle flagellomeres broader than long (Fig. 4C); process of labrum very short, about twice as broad as long, broadly truncate and very slightly emarginate medially; mandibles in repose reaching slightly beyond midline of face, with a small but distinct subapical tooth; galea quite short, impunctate, distinctly tessellate and slightly shining.

Pronotal collar without humeral angle; scutum tessellate, only slightly shining, with very obscure punctures separated by 2–4 puncture widths; scutellum tessellate,

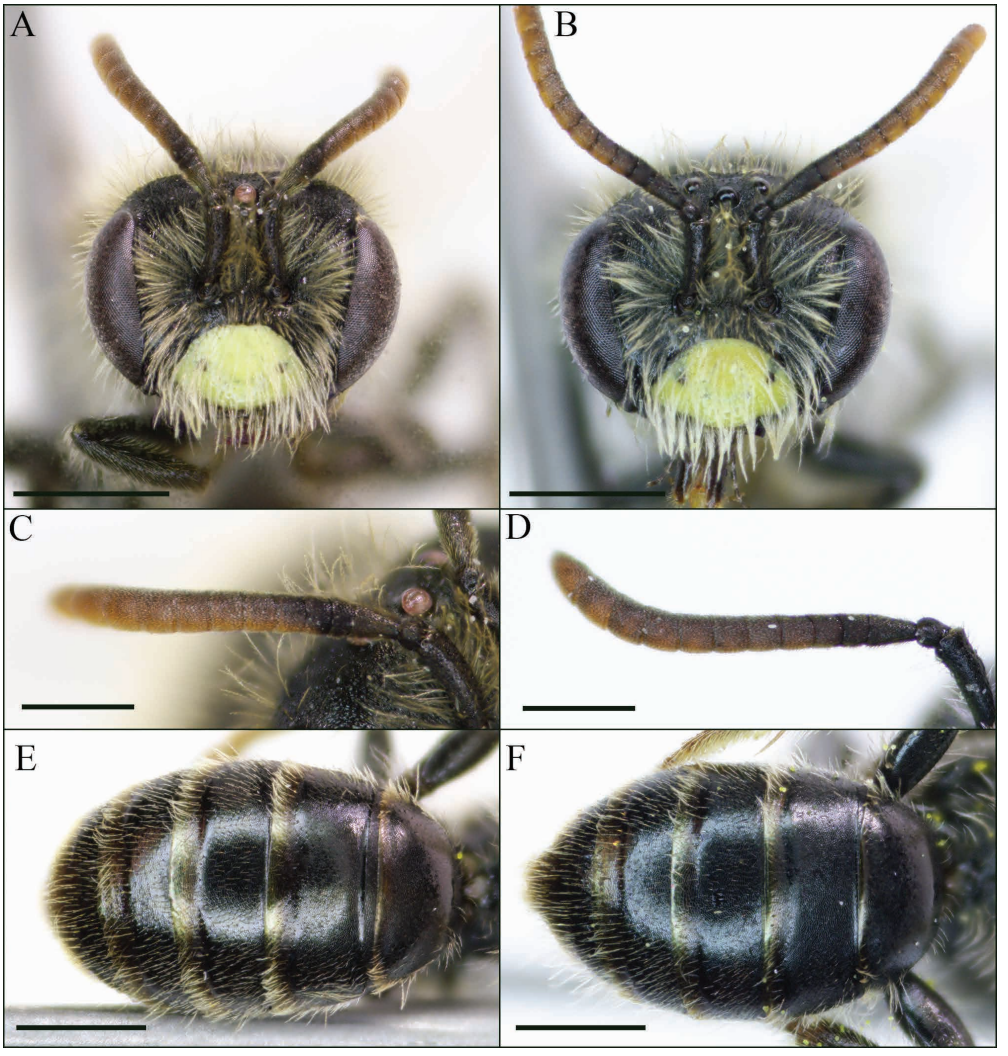


Figure 4. *Andrena vernalis* (left column) and *A. ziziae* (right column) male external characters: (A) *A. vernalis* face; (B) *A. ziziae* face; (C) *A. vernalis* antenna; (D) *A. ziziae* antenna; (E) *A. vernalis* abdomen; (F) *A. ziziae* abdomen. Scale bars: A–B, E–F = 1mm; C–D = 500 µm.

only slightly shining, with very sparse and obscure punctures; pleura granular without evident punctures; area on pleura directly anterior to mid coxae with a slightly depressed subtriangular area delimited by a slightly protruding margin (Fig. 9C–D); dorsal area of propodeum rather broad, nearly horizontal, triangle with a slightly impressed outline, finely roughened, subtly but distinctly contrasting with adjacent areas; all basitarsi slender and elongate, considerably narrower than their respective tibiae; r-rs vein relatively long with Rs vein attached to marginal cell 2–3 vein widths from the stigma; 2nd submarginal cell receiving 1m-cu vein at or slightly beyond middle.

Overall shape of metasoma ovoid, reaching greatest width at T3, equal in width to mesosoma (Fig. 4E); terga smooth and tessellate, slightly shining, punctures indistinct; T1 and T2 slightly duller than the apical terga; apical margins of terga slightly but distinctly impressed, taking up about 1/4 of segment on T2 and T3 (Fig. 4E); pygidial plate broadly triangular with a rounded apex.

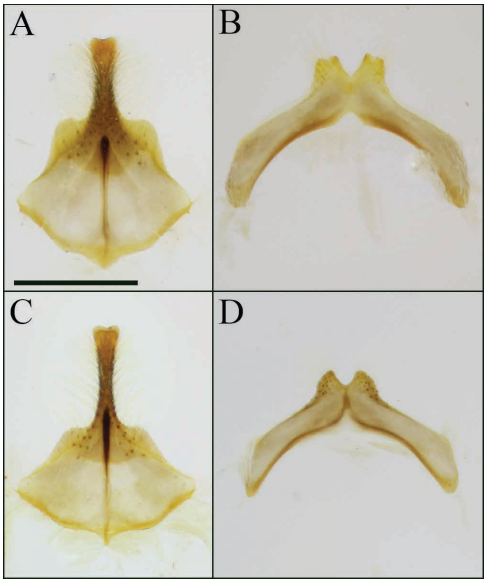


Figure 5. Male S8 and S7: *Andrena vernalis* (A) S8 and (B) S7; *A. ziziae* (C) S8 and (D) S7. Scale bar = 500 µm, all images are at the same scale.

*Terminalia.* S8 apical portion moderately slender, slightly expanded at tip (Fig. 5A); S7 with apical lobes distinctly truncate, separated by a V-shaped emargination (Fig. 5B); genitalia with dorsal lobes of gonocoxites relatively broad, narrowly separated, slightly diverging apically (Fig. 6A); ventral lobes of gonocoxides narrowed medially, apical halves expanded and strongly bent ventrally, apices slightly overlapping medially, apex of volsella appearing slightly emarginate in ventral view (Fig. 6B); penis valves quite broad medially, filling space between ventral lobes of gonocoxites.

*Vestiture.* Pubescence entirely whitish; hairs on venter of head rather long, about equal to the length of the cheek+eye; hairs on scutum erect, weakly plumose, not obscuring surface; venter of mesosoma clothed in relatively dense hairs; pubescence on discs of terga relatively short (Fig. 4E); T1 largely lacking discal pubescence; T2 discal hairs with very short and minute, discal hairs moderately longer on more apical terga; apical hair bands weak and generally worn off even in moderately worn specimens; T1 apical hair band limited to a small lateral tuft, T2 apical hair band slightly more extensive but still limited to lateral tufts, T3 apical hair band weak, diffuse, narrowly interrupted medially, T4 apical hair band entire, very weak and diffuse, T5 and T6 lacking apical hair bands (Fig. 4E).

**Description of female.** Body length: 7.2 mm, range 7.0–7.4 mm; ITD: 1.40 mm (n = 10).

See Mitchell (1960) for full description.

**Floral records. Apiaceae** (6 ♂ 108 ♀): *Zizia aptera* 6 ♂ 86 ♀, *Z. aurea* 22 ♀. Twenty-six female *A. vernalis* collected in 2018 had their body pollen analyzed. Individuals of *A. vernalis* had on average 287.5 *Zizia* pollen grains on their bodies and 97% of the pollen carried was from *Zizia* (Fig. 7).

**Phenology.** Active in May and June in Minnesota.

**Distribution.** In Minnesota, *A. vernalis* has a relatively restricted distribution, especially compared to *A. ziziae*. *Andrena vernalis* has only been found in a band in the south-central area of the state (Fig. 8A,C). Although a comprehensive examination of the range of *A. vernalis* was not performed, based on the specimens examined, it extends to the east coast of the US.

**Type material examined.** Holotype: ♀, Connecticut: Litchfield Co.: Colebrook, 31 May 1922, W.M. Wheeler leg. (MCZ 30485).

**Additional material examined.** Total specimens: 7 ♂ 114 ♀. **CONNECTICUT: Litchfield Co.:** Colebrook: 1 ♂ 2 ♀ (MCZ), 31 May 1922, W.M. Wheeler leg.; 2 ♀ (MCZ), 11 Jun 1926, W.M. Wheeler leg. **MINNESOTA: Big Stone Co.:** (45.3065 –96.2874): 1 ♀, 21 Jun 2018, S. Marconie leg., *Zizia aurea*; 2 ♀, 18 Jun 2019, I. Bur leg., *Z. aurea*; (45.3259 –96.3714): 1 ♀, 1 Jun 2018, G. Pardee leg., *Z. aptera*; 1 ♀, 1 Jun 2018, M. Rancour leg., *Z. aptera*; 15 ♀, 1 Jun 2018, S. Marconie leg., *Z. aptera*; 1 ♀, 1 Jun 2018, S. Marconie leg., *Z. aurea*; 6 ♀, 10 Jun 2019, G. Pardee leg., *Z. aptera*; 8 ♀, 10 Jun 2019, I. Bur leg., *Z. aptera*; Larson Slough WPA (45.3612 –96.3119): 1 ♀, 3 Jun 2017, P. Pennarola leg., *Z. aptera*; **Douglas Co.:** Staffanson Prairie (45.8161 –95.7460): 7 ♀, 5 Jun 2018, G. Pardee leg., *Z. aptera*; 7 ♀, 5 Jun 2018, I. Lane leg., *Z. aptera*; 1 ♀, 5 Jun 2018, T. Eicholz leg., *Z. aptera*; **Kandiyohi Co.:** (45.3529 –95.1192): 1 ♀, 25–27 May 2018, J. Brokaw leg.; 1 ♀, 5 Jun 2019, G. Pardee leg., *Z. aurea*; Brenner Lake WPA (45.4006 –95.2463): 5 ♀, 4 Jun 2018, A. Ritchie leg., *Z. aptera*; 4 ♀, 4 Jun 2018, M. Rancour leg., *Z. aptera*; 1 ♀, 7 Jun 2018, G. Pardee leg., *Z. aptera*; 6 ♀, 12 Jun 2019, G. Pardee leg., *Z. aptera*; **Lyon Co.:** Vallers WMA (44.5622 –95.8403): 1 ♀ (MNDNR), 16 Jun 2015, K.J. Jokela leg., *Z. aurea*; **Pope Co.:** (45.6707 –95.5077): 1 ♀, 3 Jun 2019, G. Pardee leg., *Z. aptera*; 2 ♂ 4 ♀, 3 Jun 2019, I. Bur leg., *Z. aptera*; 3 ♀, 11 Jun 2019, G. Pardee leg., *Z. aptera*; 3 ♀, 11 Jun 2019, I. Bur leg., *Z. aptera*; 2 ♀, 11 Jun 2019, I. Bur leg., *Z. aurea*; **Stevens Co.:** (45.4507 –96.1325): 1 ♀, 9 Jun 2017, I. Lane

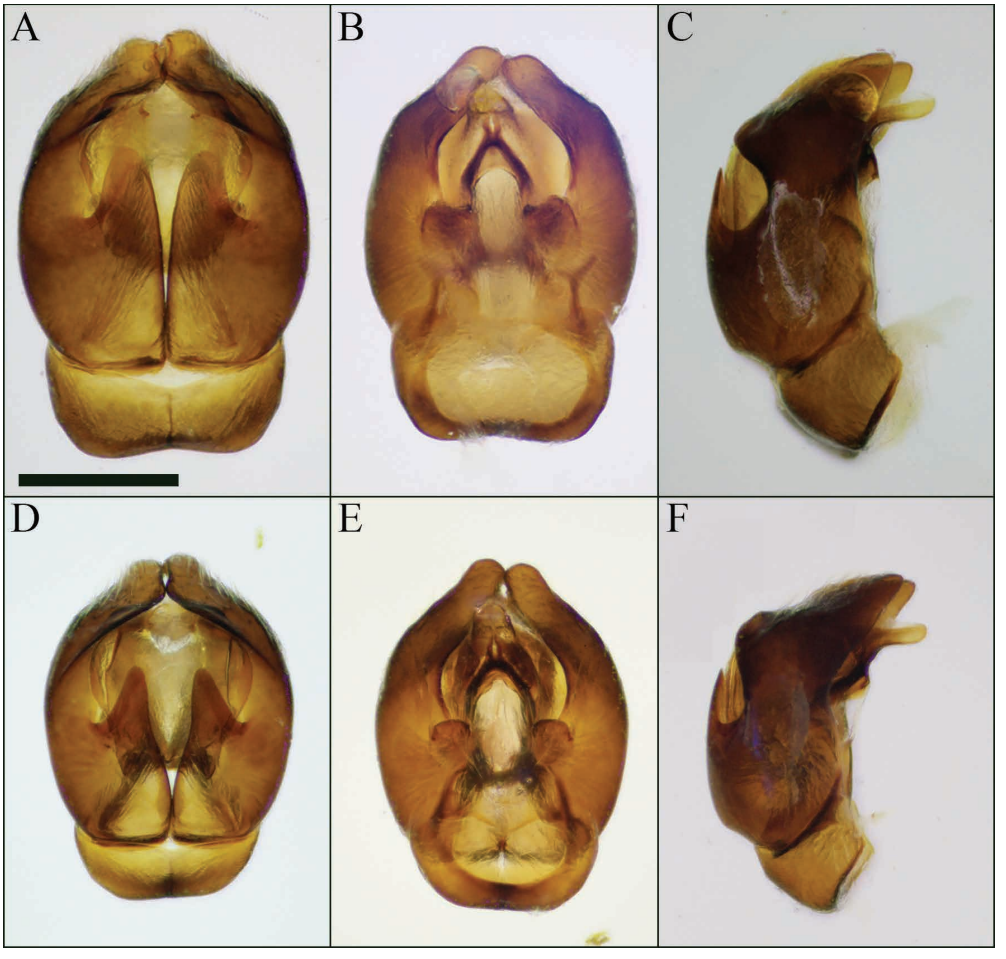


Figure 6: Male genitalia: *Andrena vernalis* (A) dorsal view; (B) ventral view; (C) lateral view; *A. ziziae* (D) dorsal view; (E) ventral view; (F) lateral view. Scale bar = 500 µm, all images are at the same scale.

leg., *Z. aurea*; 4 ♀, 7 Jun 2018, I. Lane leg., *Z. aurea*; 1 ♀, 7 Jun 2018, M. Rancour leg., *Z. aurea*; 3 ♀, 7 Jun 2018, S. Marconie leg., *Z. aurea*; 3 ♀, 7 Jun 2018, T. Eicholz leg., *Z. aurea*; 1 ♀, 18 Jun 2019, I. Bur leg., *Z. aurea*; John Freeman WMA (45.4611 –95.9681): 5 ♀, 6 Jun 2019, G. Pardee leg., *Z. aptera*; 2 ♀, 6 Jun 2019, I. Bur leg., *Z. aptera*; **Swift Co.:** Chippewa Prairie (45.1545 –96.0086): 1 ♀, 31 May 2018, A. Ritchie leg., *Z. aptera*; 4 ♂ 4 ♀, 31 May 2018, I. Lane leg., *Z. aptera*; **Washington Co.:** Belwin Conservancy (44.9445 –92.8169): 1 ♀, 7 Jun 2016, E. Evans leg., *Z. aurea*. **VERMONT: Franklin Co.:** St. Albans: 1 ♀ (MCZ), 21 Jun 1913, W.M. Wheeler leg.

**Remarks.** Ribble (1968) synonymized *A. vernalis* with *A. ziziae* because he considered *A. vernalis* to merely represent variation within *A. ziziae*. One of the defining characters that Mitchell (1960) used to separate *A. vernalis* was the pleural depression and ridge; Ribble (1968) considered this character too variable, stating:

"Different individuals show a continuous variation in the mesepisternum from the modified type (above) to specimens having a flattened area in front of the coxa. Also, individuals with modified mesepisterna occur throughout most of the range of *ziziae* and are often collected with it. Specimens collected together may be almost identical except for the quite different mesepisterna and intermediates between these two types may occur with them. Males occasionally have weakly depressed areas in front of the middle coxae, but are not unusual in other respects."

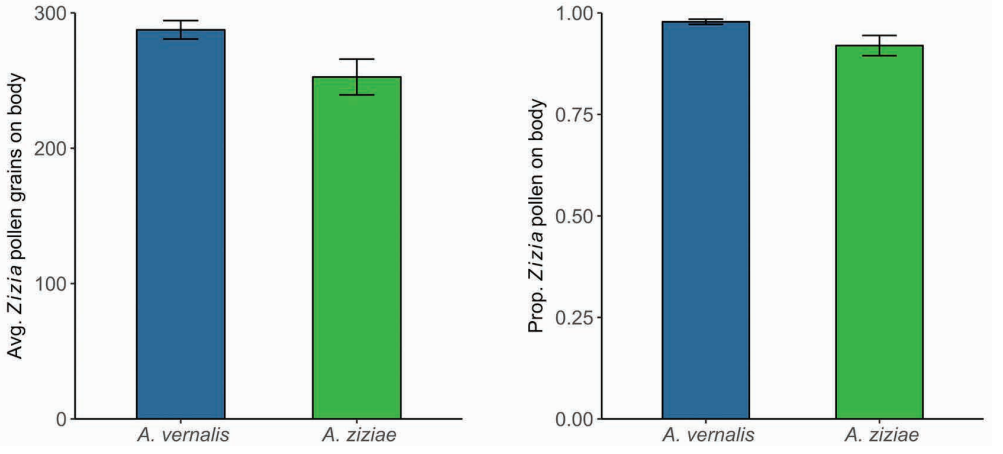


Figure 7. Foraging behavior of *A. vernalis* (blue) and *Andrena ziziae* (green) based on our pollen study. The left panel shows the average number of *Zizia* pollen grains found on individuals of each species and the right panel shows the average proportion of *Zizia* pollen grains found on individuals of each species. Bars are means ± SE.

However, Ribble (1968) followed that with a discussion on the variation in the width of the facial fovea in the female and the shape of the apical lobes of the male S7. Both of those characters are generally consistent within species and are important in delineating species boundaries.

Ribble (1968) did not examine the holotype of *A. vernalis*, though he appears to have examined the two paratypes at NCSU. A note on the location of the paratypes: although the holotype of *A. vernalis* is located at MCZ and Mitchell (1960) indicated that three of the four paratypes were also deposited at MCZ, none of the paratypes were able to be located there. Two of them (from Colebrook, CT) are currently at NCSU and it's not clear where the third purported MCZ paratype (collected from Pine Island, NY) is located.

Examination of specimens reveals that in addition to the pleural depression and ridge, there are multiple other consistent characters that differentiate *A. vernalis* and *A. ziziae*, including the width of the facial fovea, sculpturing of the propodeum, length of tergal hair bands, length of transparent apical rims, length of the r-rs crossvein, relative length of the antennal segments, and the male terminalia. These characters are discussed in depth in the diagnosis for *A. vernalis*. The pleural ridge is indeed one of the more variable characters, ranging from deep with a strong carina to shallow with a weak carina in *A. vernalis*. The pleural depression and ridge are weaker in the males than the females, though it is consistently present. Some *A. ziziae* females have a weak pleural depression, but it never reaches the level seen in *A. vernalis* and no specimens were found that fully intergrade. Overall the pleural depression and ridge is a consistent character that can distinguish *A. vernalis*, but it is often partially or fully obscured by the midlegs and wings, requiring it to be viewed at a non-ideal angle if it is even visible at all. As a result, in females, the facial fovea is the clearest and most consistent defining character, though the difference is difficult or impossible to see if the foveae are matted.

The species status of *A. vernalis* is most strongly supported by the differences in male terminalia (see diagnosis for details). Six male *A. vernalis* were collected at the same collection events as females and one additional male was found at MCZ that was collected at the same collection event as the holotype but was apparently never seen by Mitchell. The males and females of *A. vernalis* are associated based on the shared pleural depression and ridge, the relatively shorter antennae in both sexes, and the relatively longer clear apical rims of the terga. Plus, the correct association between the male and female in *A. ziziae* is clear, since the lectotype of *A. ziziae* was caught in copula and pinned with the male. In addition, numerous male *A. ziziae* were collected in association with females (including two in copula) at sites where *A. vernalis* was not found. In light of these clear and consistent morphological differences in both males and females, we are reinstating *A. vernalis* as a valid species.

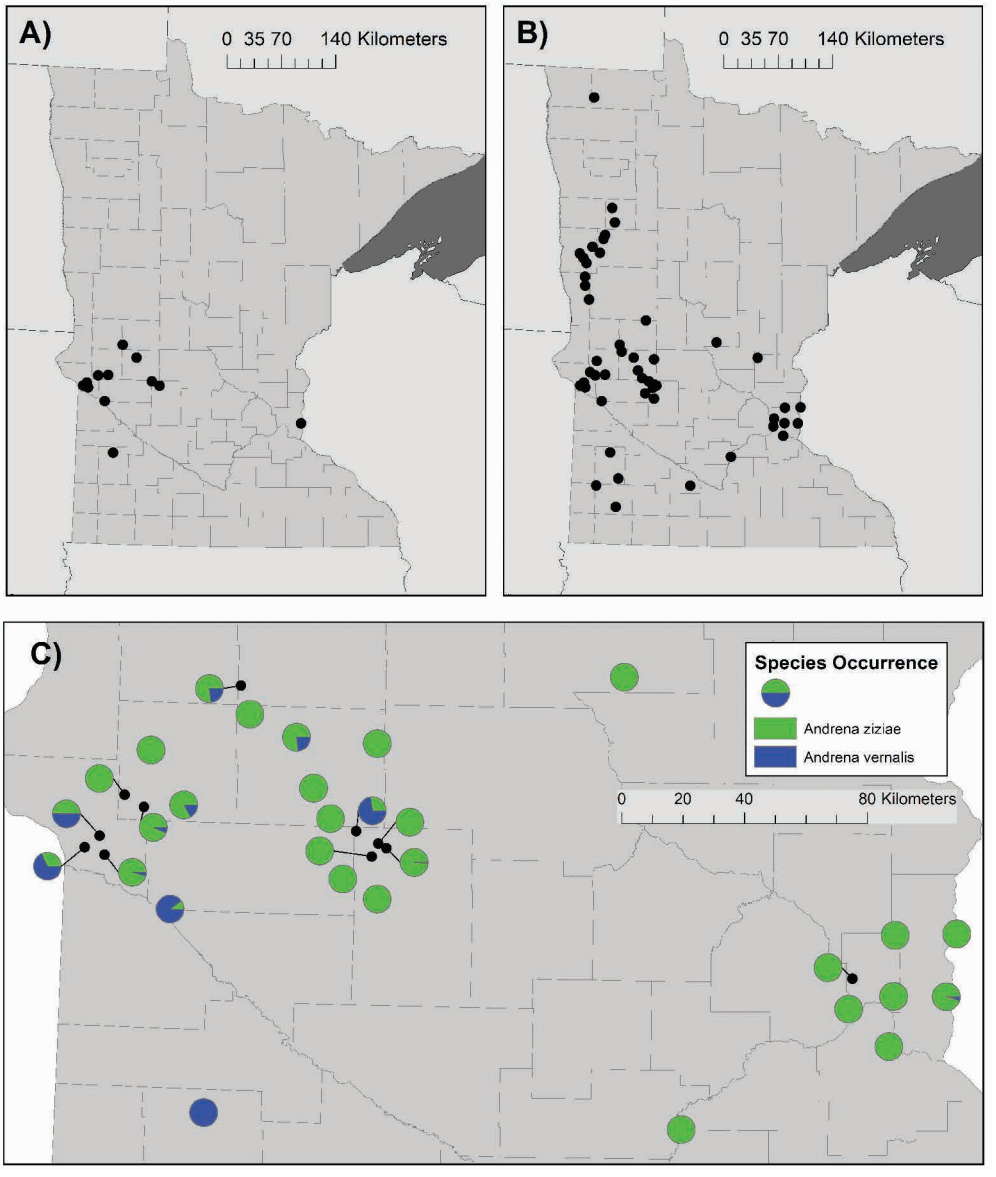


Figure 8. Distribution of *Andrena vernalis* and *A. ziziae* in Minnesota: (A) distribution of *A. vernalis*; (B) distribution of *A. ziziae*; (C) area where the range of *A. vernalis* and *A. ziziae* overlap, showing the relative proportion of each species at each site.

Examination of specimens at MCZ revealed five additional topotypical specimens of *A. vernalis* (4 ♀ and 1 ♂), with two females and the male collected from the same collection event as the type. All the specimens have a prominent pleural depression and ridge (Fig. 9). Indeed, this character seems to be enhanced because the faded integument of these aged specimens provides better contrast. In addition to the pleural depression and ridge, the male and female topotypical specimens and the holotype are a morphological match with the specimens from Minnesota. As a result, we can confidently assign the Minnesota specimens to *A. vernalis* despite the moderate geographic distance between them.

Historically, there has been a fair amount of confusion regarding the identity

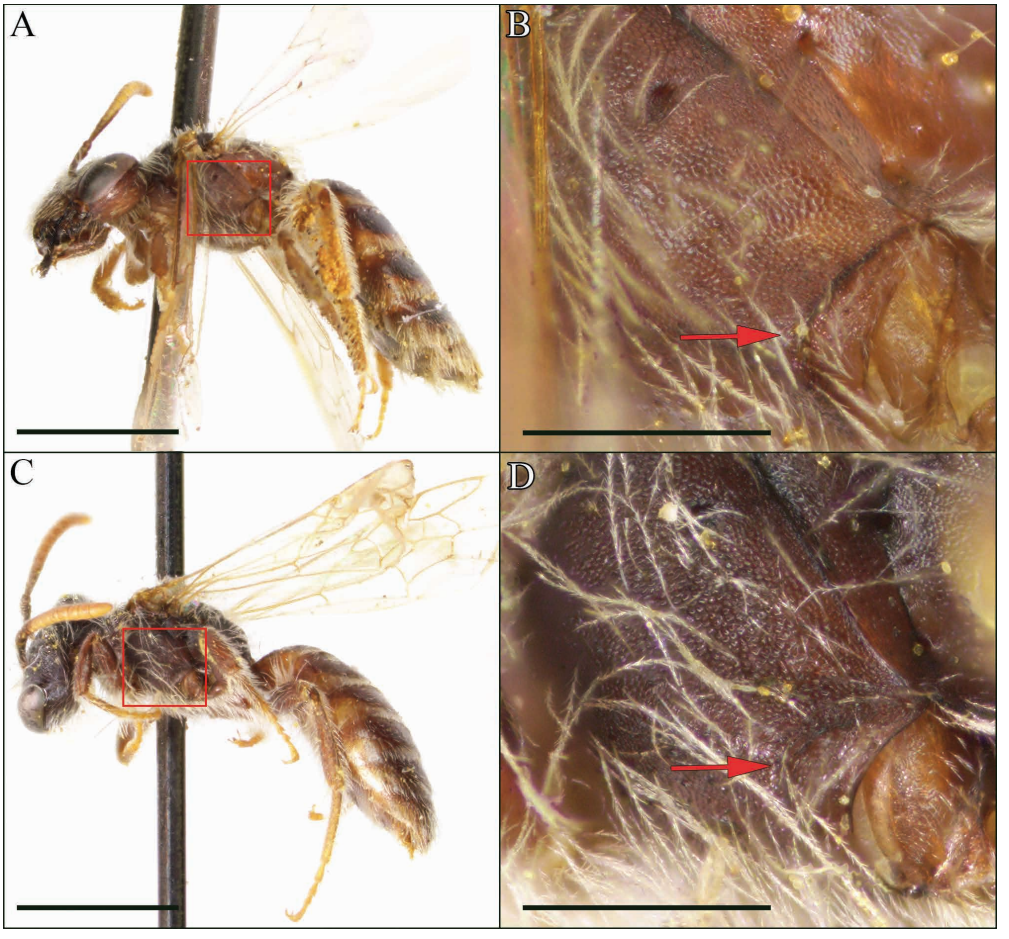


Figure 9. Topotypical specimens of *A. vernalis* with pleural depression and ridge: (A) female lateral view; (B) closeup of female pleural depression and ridge, indicated by red arrow; (C) male lateral view; (D) closeup of male pleural depression and ridge, indicated by red arrow. Scale bars: A–B = 2 mm; C–D = 500 µm.

of *A. vernalis*. Even Mitchell (1960), despite originally describing *A. vernalis*, may have confused some specimens with *A. ziziae*, since he noted that some specimens of *A. ziziae* had broader facial fovea and more broadly hyaline apical impressed areas of the abdominal terga; both of these characters are suggestive of *A. vernalis*. The terminalia illustrations by Ribble (1968) for *A. ziziae* do not match the current understanding of that species; instead, the illustrations match our current understanding of *A. vernalis*. In contrast, the terminalia illustrations of *A. ziziae* by Mitchell (1960) are consistent with our current understanding of *A. ziziae*. Finally, Ribble (1968) extensively discussed the apparent variation in *A. ziziae*, including variation in the width of the fovea, the shape of the male S7, and pleural ridge.

The *A. ziziae*/*vernalis* complex has sometimes been confused with other species. For example, one female *A. vernalis* was found misidentified as *A. personata* in the MCZ collection (identifier unknown) and a mix of females (6 *A. ziziae* and 1 *A. vernalis*) from Minnesota were all misidentified as *A.* (*Simandrena*) *nasonii* Robertson, 1895 by S. Droege.

***Andrena* (*Micrandrena*) *ziziae* Robertson**

Fig. 1B,D,F, 2B,D,F, 3B,D,F, 4B,D,F, 5C–D, 6D–F, 7, 8B–C

*Andrena ziziae* Robertson, 1891: 55, ♀♂ (in part). Lectotype ♀ (designated by Ribble 1968), USA, IL, Carlinville, 7 May 1887, C. Robertson leg., on *Zizia aurea*, INHS #179494; Robertson, 1897: 335 (re-

description, separation from *A. personata*); Bruner, 1903: 242 (key); Viereck, 1916: 712, 716 (key).

*Opandrena ziziae*: Robertson, 1902: 193 (new generic assignment, key).

*Micrandrena ziziae*: Cockerell, 1909: 420 (new generic assignment).

*Andrena* (*Micrandrena*) *ziziae*: Cockerell, 1932: 157–158 (subgeneric assignment, key); Mitchell, 1960: 93, 99, 168–169, Fig. 31, 32 (key, redescription); Ribble, 1968, 246, 267 (key, redescription) (in part); Hurd 1979: 1810 (catalogue).

**Diagnosis.** See diagnosis of *A. vernalis* in order to separate these two species.

Both females and males key out correctly in Mitchell (1960); since male *A. vernalis* are not included in Mitchell (1960), see diagnosis of *A. vernalis* to separate males.

*Andrena ziziae* males key out to the *A. ziziae/piperi* Viereck couplet in Ribble (1968) but don't quite match the *A. ziziae* couplet since its antennal segments are longer than would be expected by the key. Females key out to the A. *ziziae/chlorogaster* Viereck couplet in Ribble (1968) but the fovea are slightly narrower than would be expected by the key (taking up slightly more than half the space between the eye and lateral ocelli). Since neither male nor female *A. vernalis* are included in Ribble (1968), see the diagnosis of *A. vernalis* to separate from *A. ziziae*.

**Description of male.** Body length: 6.0 mm, range 5.5–6.3 mm; ITD: 1.22 mm (n = 10).

See Mitchell (1960) for full description.

**Description of female.** Body length: 6.9 mm, range 6.5–7.3 mm; ITD: 1.33 mm (n = 10).

See Mitchell (1960) for full description.

**Floral records. Apiaceae** (58 ♂ 642 ♀): *Zizia aptera* 5 ♂ 151 ♀, *Z. aurea* 53 ♂ 491 ♀, **Asteraceae**: *Taraxacum officinale* 3 ♂ 6 ♀, **Fabaceae**: *Trifolium repens* 1 ♀. Thirty-five female *A. ziziae* collected in 2018 had their body pollen analyzed. Individuals of *A. ziziae* had on average 252 *Zizia* pollen grains on their bodies and 91.95% of the pollen carried was from *Zizia* (Fig. 7).

**Phenology.** Active in May and June in Minnesota.

**Type material examined.** Lectotype ♀, Illinois: Macoupin Co., Carlinville, 7 May 1887, C. Robertson leg., on *Z. aurea*. Robertson #3819, INHS #179,494 (INHS). 1 Paralectotype, ♂, on same pin as lectotype (INHS).

**Additional material examined.** Total specimens: 61 ♂ 664 ♀. **CONNECTICUT: Litchfield Co.:** Colebrook: 2 ♀ (MCZ), 8–11 Jun 1911, W.M. Wheeler leg.; 5 ♀ (MCZ), 31 May 1922, W.M. Wheeler leg. **MINNESOTA: Becker Co.:** (47.0337 –96.0814): 2 ♀, 8 Jun 2017, R. Tucker leg., *Zizia aurea*; Heliksen WPA (47.0793 –96.0579): 2 ♂, 8 Jun 2017, C. Herron-Sweet leg., *Z. aptera*; 2 ♀, 8 Jun 2017, C. Herron-Sweet leg., *Z. aurea*; 3 ♀, 27 Jun 2017, A. Waananen leg., *Z. aurea*; Lunde WMA (46.8710 –96.1321): 1 ♀, 13 Jun 2018, D. Drons leg., *Z. aurea*; Lunde WMA (46.8713 –96.1319): 3 ♀ (2 ♀ MNDNR), 13 Jun 2018, D. Drons leg., *Z. aurea*; **Big Stone Co.:** (45.3065 –96.2874): 6 ♀, 9 Jun 2017, C. Herron-Sweet leg., *Z. aurea*; 2 ♀, 1 Jun 2018, A. Ritchie leg., *Taraxacum officinale*; 4 ♀, 1 Jun 2018, A. Ritchie leg., *Z. aurea*; 5 ♀, 1 Jun 2018, I. Lane leg., *Z. aurea*; 9 ♀, 1 Jun 2018, T. Eicholz leg., *Z. aurea*; 9 ♀, 21 Jun 2018, S. Marconie leg., *Z. aurea*; 9 ♀, 18 Jun 2019, G. Pardee leg., *Z. aurea*; 22 ♀, 18 Jun 2019, I. Bur leg., *Z. aurea*; (45.3259 –96.3714): 2 ♀, 1 Jun 2018, S. Marconie leg., *Z. aptera*; 8 ♀, 10 Jun 2019, G. Pardee leg., *Z. aptera*; 5 ♀, 10 Jun 2019, I. Bur leg., *Z. aptera*; Larson Slough WPA (45.3612 –96.3119): 1 ♀, 3 Jun 2017, P. Pennarola leg., *Z. aptera*; **Brown Co.:** Joseph A. Tauer Prairie SNA (44.2009 –94.5326): 1 ♀, 18 Jun 2010, C. Kern leg.; **Clay Co.:** (46.8016 –96.4056): 1 ♂, 31 May 2017, C. Herron-Sweet leg., *Z. aurea*; 1 ♀, 22 Jun 2017, R. Tucker leg., *Z. aurea*; Bluestem SNA (46.8542 –96.4723): 1 ♀, 31 May 2017, R. Tucker leg., *Z. aurea*; 1 ♀, 22 Jun 2017, C. Herron-Sweet leg., *Z. aurea*; Clay County WMA (46.7478 –96.3535): 5 ♀, 19 Jun 2017, A. Waananen leg., *Z. aurea*; 8 ♀, 19 Jun 2017, R. Tucker leg., *Z. aurea*; Hoykens WPA (46.9368 –96.2631): 2 ♂ 8 ♀, 6 Jun 2017, R. Tucker leg., *Z. aurea*; **Dakota Co.:** Pine Bend Bluffs SNA (44.7912 –93.0320): 1 ♀, 4 Jun 2013, C. Boyd leg., *Z. aurea*; **Douglas Co.:** Miltona WMA (46.1073 –95.3218): 2 ♀, 13 Jun 2018, L. Gedlinske leg., *Z. aurea*; Staffanson Prairie (45.8161 –95.7460): 28 ♀, 5 Jun 2018, G. Pardee leg., *Z. aptera*; 15 ♀, 5 Jun 2018, I. Lane leg., *Z. aptera*; 7 ♀, 5 Jun 2018, T. Eicholz leg., *Z. aptera*; **Isanti Co.:** Dalbo WMA (45.6962 –93.4558): 1 ♀ (MNDNR), 20 Jun 2018, N. Gerjets leg., *Z. aurea*; **Kandiyohi Co.:** (45.2031 –95.1528): 1 ♂, 25 May 2017, R. Tucker leg., *Z. aurea*; (45.3273 –95.1790): 5 ♀, 2 Jun 2017, I. Lane leg., *Z. aurea*; (45.3529 –95.1192): 21 ♀, 2 Jun 2017, C. Herron-Sweet leg., *Z. aurea*; 1 ♀, 4–6 Jun 2018, J. Brokaw leg.; 12 ♀, 4 Jun 2018, I. Lane leg., *Z. aurea*; 15 ♀, 4 Jun 2018, S. Marconie leg., *Z. aurea*; 19 ♀, 4 Jun 2018, T. Eicholz leg., *Z. aurea*; 1 ♀, 27 Jun 2018, G. Pardee leg., *Z. aurea*; 2 ♂ 18 ♀, 5 Jun 2019, G. Pardee leg., *Z. aurea*; 4 ♂ 18 ♀, 5 Jun 2019, I. Bur leg., *Z. aurea*; 5 ♀, 12 Jun 2019, G. Pardee leg., *Z. aurea*; 5 ♀, 12 Jun 2019,

I. Bur leg., *Z. aurea*; (45.3658 –95.1537): 2 ♀, 18 Jun 2019, B. Bruninga-Socolar, M. Dutta, D. Harder leg.; Brenner Lake WPA (45.4006 –95.2463): 3 ♀, 4 Jun 2018, A. Ritchie leg., *Z. aptera*; 1 ♀, 12 Jun 2019, G. Pardee leg., *Z. aptera*; 2 ♀, 12 Jun 2019, I. Bur leg., *Z. aptera*; **Lyon Co.:** Glynn Prairie SNA (44.2638 –95.6962): 1 ♀, 14 May 2017, P. Pennarola leg., *Z. aptera*; **Mahnomen Co.:** Santwire WMA (47.2303 –95.8998): 1 ♀ (MNDNR), 19 Jun 2018, D. Drons leg., *Z. aurea*; Wambach WMA (47.3976 –95.9536): 1 ♀, 6 Jun 2018, D. Drons leg., *Z. aurea*; **Morrison Co.:** Rice Area Sportsmens Club WMA (45.8685 –94.1419): 2 ♀, 12 Jun 2018, L. Gedlinske leg., *Z. aurea*; **Murray Co.:** Lundblad Prairie SNA (43.9347 –95.7197): 1 ♀, 8 Jun 2015, K.J. Jokela leg., *Z. aurea*; Ruthton WMA (44.1729 –96.0463): 3 ♀, 15 Jun 2015, A. Fulton leg.; **Pope Co.:** (45.4353 –95.3556): 1 ♀, 14 Jun 2017, R. Tucker leg., *Z. aurea*; (45.5229 –95.4303): 13 ♂ 6 ♀, 31 May 2017, I. Lane leg., *Z. aurea*; (45.6707 –95.5077): 1 ♂ 5 ♀, 3 Jun 2019, G. Pardee leg., *Z. aptera*; 2 ♂ 14 ♀, 3 Jun 2019, I. Bur leg., *Z. aptera*; 17 ♀, 11 Jun 2019, G. Pardee leg., *Z. aptera*; 5 ♀, 11 Jun 2019, I. Bur leg., *Z. aptera*; 5 ♀, 11 Jun 2019, I. Bur leg., *Z. aptera*; 5 ♀, 11 Jun 2019, I. Bur leg., *Z. aurea*; (45.7357 –95.7054): 2 ♂ 5 ♀, 5 Jun 2018, A. Ritchie leg., *Z. aurea*; 1 ♂, 5 Jun 2018, M. Rancour leg., *Z. aurea*; 8 ♀, 5 Jun 2018, S. Marconie leg., *Z. aurea*; 2 ♀, 22 Jun 2018, T. Eicholz leg., *Z. aurea*; 3 ♂ 4 ♀, 4 Jun 2019, G. Pardee leg., *Z. aurea*; 1 ♂ 6 ♀, 4 Jun 2019, I. Bur leg., *Z. aurea*; Krantz Lake WPA (45.6590 –95.1701): 2 ♀, 20 Jun 2018, L. Gedlinske leg., *Z. aurea*; Krantz Lake WPA (45.6591 –95.1693): 1 ♀, 20 Jun 2018, L. Gedlinske leg., *Z. aurea*; Krantz Lake WPA (45.6592 –95.1691): 1 ♀, 20 Jun 2018, L. Gedlinske leg., *Z. aurea*; Krantz Lake WPA (45.6596 –95.1692): 1 ♀, 20 Jun 2018, L. Gedlinske leg., *Z. aurea*; Krantz Lake WPA (45.6650 –95.1684): 1 ♂ (MNDNR), 31 May 2018, N. Gerjets, L. Gedlinske leg., *Z. aurea*; Krantz Lake WPA (45.6651 –95.1684): 1 ♂ (MNDNR), 31 May 2018, N. Gerjets, L. Gedlinske leg., *Z. aurea*; Krantz Lake WPA (45.6653 –95.1679): 1 ♂ 1 ♀ (MNDNR), 31 May 2018, N. Gerjets, L. Gedlinske leg., *Z. aurea*; Krantz Lake WPA (45.6653 –95.1682): 1 ♂ (MNDNR), 31 May 2018, N. Gerjets, L. Gedlinske leg., *Z. aurea*; Krantz Lake WPA (45.6654 –95.1680): 1 ♀, 31 May 2018, N. Gerjets, L. Gedlinske leg., *Z. aurea*; **Ramsey Co.:** Bald Eagle-Otter Lake Regional Park (45.1171 –93.0059): 7 ♀, 5 Jun 2015, E. Evans leg., *Z. aurea*; Battle Creek Regional Park (44.9380 –93.0126): 1 ♀, 8 Jun 2016, J. Gardner leg., *Trifolium repens*; 3 ♀, 8 Jun 2016, J. Gardner leg., *Z. aurea*; Battle Creek Regional Park (44.9380 –93.0127): 4 ♂ 4 ♀, 27 May 2015, J. Gardner leg., *Z. aurea*; Battle Creek Regional Park (44.9386 –92.9914): 4 ♀, 10 Jun 2015, J. Gardner leg., *Z. aurea*; Battle Creek Regional Park (44.9394 –92.9881): 2 ♀, 8 Jun 2016, J. Gardner leg., *Z. aurea*; UMN Bee Lab Garden (44.9893 –93.1815): 2 ♂ 1 ♀, 28 May 2019, I. Bur leg., *Z. aurea*; 1 ♀, 31 May 2019, I. Bur leg., *Z. aptera*; **Roseau Co.:** Two Rivers Aspen Parkland SNA (48.6679 –96.3444): 1 ♂ (MNDNR), 30 May 2015, C. Boyd leg., *Z. aurea*; **Scott Co.:** Ney WMA (44.5445 –93.8825): 1 ♀, 14 Jun 2018, N. Gerjets leg., *Z. aurea*; Ney WMA (44.5446 –93.8825): 1 ♀, 14 Jun 2018, N. Gerjets leg., *Z. aurea*; Ney WMA (44.5447 –93.8822): 1 ♀ (MNDNR), 14 Jun 2018, N. Gerjets leg., *Z. aurea*; **Stevens Co.:** (45.4507 –96.1325): 3 ♀, 9 Jun 2017, A. Waananen leg., *Z. aurea*; 1 ♂ 28 ♀, 9 Jun 2017, I. Lane leg., *Z. aurea*; 3 ♂ 4 ♀, 22 May 2018, I. Lane leg., *T. officinale*; 1 ♂, 22 May 2018, I. Lane leg., *Z. aurea*; 2 ♂ 23 ♀, 7 Jun 2018, I. Lane leg., *Z. aurea*; 16 ♀, 7 Jun 2018, M. Rancour leg., *Z. aurea*; 3 ♂ 60 ♀, 7 Jun 2018, S. Marconie leg., *Z. aurea*; 1 ♂ 25 ♀, 7 Jun 2018, T. Eicholz leg., *Z. aurea*; 5 ♀, 18 Jun 2019, G. Pardee leg., *Z. aurea*; 7 ♀, 18 Jun 2019, I. Bur leg., *Z. aurea*; (45.4841 –96.2151): 2 ♀, 9 Jun 2017, R. Tucker leg., *Z. aurea*; Cin WMA (45.6184 –96.1130): 1 ♀, 18 Jun 2015, K.J. Jokela leg., *Z. aurea*; John Freeman WMA (45.4611 –95.9681): 1 ♀, 22 May 2018, A. Ritchie leg., *Z. aptera*; 15 ♀, 6 Jun 2019, G. Pardee leg., *Z. aptera*; 19 ♀, 6 Jun 2019, I. Bur leg., *Z. aptera*; **Swift Co.:** Bengtson WPA (45.2593 –95.2977): 3 ♀, 1 Jun 2017, P. Pennarola leg., *Z. aurea*; Chippewa Prairie (45.1545 –96.0086): 1 ♀, 31 May 2018, I. Lane leg., *Z. aptera*; **Washington Co.:** Coldwater Spring (44.9007 –93.1977): 1 ♂, 31 May 2017, K. Friedrich leg., *Z. aurea*; Arcola Bluffs (45.1210 –92.7510): 1 ♀, 7 Jun 2017, K. Friedrich leg., *Z. aurea*; Belwin Conservancy (44.9367 –92.7952): 1 ♂, 18 May 2016, J. Gardner leg., *Z. aurea*; Belwin Conservancy (44.9444 –92.8169): 2 ♀, 9 Jun 2015, E. Evans leg., *Z. aurea*; Belwin Conservancy (44.9445 –92.8169): 11 ♀, 7 Jun 2016, E. Evans leg., *Z. aurea*; Belwin Conservancy (44.9450 –92.8170): 1 ♀, 18 May 2016, J. Gardner leg.; Belwin Conservancy (44.9455 –92.8175): 3 ♀, 9 Jun 2015, E. Evans leg., *Z. aurea*; Belwin Conservancy (44.9482 –92.7853): 2 ♂, 9 Jun 2015, E. Evans leg., *Z. aurea*; **Wilkin Co.:** Foxhome Prairie Preserve TNC (46.3246 –96.2825): 2 ♀, 21 Jun 2018, L. Gedlinske leg., *Z. aurea*; Rice SNA (46.5884 –96.3680): 3 ♀, 20 Jun 2017, R. Tucker leg., *Z. aurea*; Tanberg 29-1 (46.4838 –96.3569): 1 ♀, 9 Jun 2017, P. Pennarola leg., *Z. aurea*.

**Remarks.** Examination of the lectotype and one of the paralectotypes reveal that *A. ziziae* match the traditional definition of that species (i.e. does not match *A. vernalis*). As noted by Ribble (1968), some

male *A. ziziae* have small yellow spots on the paraocular areas. One female was found with 2 submarginal cells on one wing and the normal 3 on the other.

Note that the terminalia illustrations by Mitchell (1960, Fig. 31, 32) are correct, whereas the terminalia illustrations by Ribble (1968, Fig. 26–29) actually match *A. vernalis*. The illustration of S8 by Ribble (1968, Fig. 30) lacks key details and could potentially match either *A. ziziae* or *A. vernalis*.

## Discussion

Here, we reinstated *A. vernalis*, which had been previously synonymized with *A. ziziae* by Ribble (1968). Although the taxonomic treatment of a single species in a limited range is not ideal, we believe it is warranted in this case because we are reinstating a synonym rather than describing an entirely new species. In addition, a clear species concept of *A. vernalis* is needed for upcoming pollinator studies on Minnesota prairies where *A. vernalis* features prominently. Finally, the apparently restricted habitat requirements of *A. vernalis* in Minnesota make it a potential species of conservation concern.

It is our hope that the identification resources provided here will allow other researchers to confidently separate *A. vernalis* and *A. ziziae* and flesh out the respective ranges of these two species. The frequent co-occurrence of these two similar species is particularly intriguing. In general, *A. ziziae* was more widely distributed and more abundant than *A. vernalis* across Minnesota, though there was substantial overlap between the two species (Fig. 8). Of the 50 locations in our study where *A. ziziae* was found, *A. vernalis* was also present at 12 of them (Fig. 8C). There was only a single site where *A. vernalis* was found but *A. ziziae* was not, and this was represented by a single specimen. This co-occurrence pattern may hold throughout the range of these two species, since their frequent co-occurrence was a primary reason that Ribble (1968) synonymized *A. vernalis* with *A. ziziae*, and specimens of *A. vernalis* and *A. ziziae* even co-occurred in the type locality of *A. vernalis* (Colebrook, CT).

Sites with *A. vernalis* were located exclusively in central Minnesota, where six sites had greater than seven individuals captured. Of these sites, five of them were considered high quality remnant prairie, while the sixth was an older prairie restoration (> 6 years). These six sites were all topographically complex, containing both wet lowlands and well-drained uplands. The species of *Zizia* differed somewhat between prairie remnants and restorations, with *Zizia aurea* more characteristic of restorations, and *Z. aptera* more characteristic of prairie remnants. In general, *A. vernalis* was much more commonly found on *Z. aptera* than on *Z. aurea*, though it is unclear if this constitutes a floral preference or is a function *of Z. aptera* dominating the high-quality remnants where *A. vernalis* was most commonly found.

Regardless of habitat type, *A. vernalis* and *A. ziziae* were strongly associated with *Zizia*. The vast majority of specimens were collected through hand netting from *Zizia* spp., though a small number were netted from other plant species (n = 10) or were collected through passive traps (n = 5). The goal of one study that provided 480 specimens for this work was to characterize entire bee community floral associations, meaning that netting was performed on all flowering plants in the system throughout the flowering season (Lane et al. 2020, I. Lane, unpublished data). However, despite this whole-community sampling, all *A. vernalis* were caught on *Zizia* and only nine *A. ziziae* were caught on a non-*Zizia* host (*Taraxacum*). In addition, the high fidelity of *A. vernalis* and *A. ziziae* to *Zizia* is supported by the high proportion of *Zizia* pollen found on the bodies of both species (Fig. 7). This suggests that *Zizia* is the primary nectar source as well as pollen source for both bee species. While *Zizia* are by far the dominant spring plants in most of the systems where collection took place for this study, it remains unknown to what extent these two species also use the pollen of other Apiaceae. However, in Minnesota, related species of Apiaceae typically do not share the same habitat or flowering phenology as *Zizia*.

Finally, reinstating *A. vernalis* creates some uncertainty on previous studies that recorded *A. ziziae*. For example, previous studies on the pollination of *Zizia* and *Thaspium* (Lindsey 1984, Lindsey and Bell 1985) may have a more complex system that initially supposed if *A. ziziae* and *A. vernalis* both occur in those systems. In addition, once both species are taken into account, previous work showing that *A. ziziae* is broadly oligolectic on Apiaceae may reveal that one or both of the species have even more specialized floral preferences (Robertson 1926, Ribble 1968, LaBerge 1986, Wood and Roberts 2018). In addition, faunal studies and checklists that recorded *A. ziziae* (e.g. Wolf and Ascher 2008, Smith et al. 2012, Harmon-Threatt and Hendrix 2015, Gibbs et al. 2017) should be revisited in order to determine which species are present.

**Acknowledgments**

We would like to thank the many people who generously provided specimens and data used in this study: Julia Brokaw (UMN), Bethanne Bruninga-Socolar (UMN), Elaine Evans (UMN), Kiley Friedrich (UMN), Nicole Gerjets (MNDNR), Nora Pennarola (UMN), and Jessica Petersen (MNDNR). This work was funded by the Minnesota Environment and Natural Resources Trust Fund, M.L. 2016, Chp. 186, Sec. 2, Subd. 03a. Specimens collected by MNDNR were also funded by the Minnesota Environment and Natural Resources Trust Fund. We also appreciate the assistance of the curators who provided access to type specimens or data: Crystal Maier (MCZ), Thomas McElrath (INHS), and Bob Blinn (NCSU). We thank Corey Sheffield and one anonymous reviewer for their helpful comments that improved this manuscript. Finally, we would like to thank Mike Arduser for providing feedback on the characters used to separate *A. vernalis* and *A. ziziae*.

**Literature Cited**

**Bruner, L. 1903**. Some notes on the bee genus *Andrena*. Transactions of the American Entomological Society. 29: 239–258.

**Cockerell, T. D. A. 1909**. Descriptions of some bees in the U.S. National Museum. Proceedings of the United States National Museum. 36: 411–420.

**Cockerell, T. D. A. 1932**. Two new bees of the genus *Andrena* from Canada. Canadian Entomologist. 64: 155–158.

**Davis, J. D., S. D. Hendrix, D. M. Debinski, and C. J. Hemsley**. **2008**. Butterfly, bee and forb community composition and cross-taxon incongruence in tallgrass prairie fragments. Journal of Insect Conservation. 12: 69–79.

**Engel, M. S. 2001**. A monograph of the Baltic Amber bees and evolution of the Apoidea (Hymenoptera). Bulletin of the American Museum of Natural History. 259: 1–192.

**Gibbs, J., J. S. Ascher, M. G. Rightmyer, and R. Isaacs**. **2017**. The bees of Michigan (Hymenoptera: Apoidea: Anthophila), with notes on distribution, taxonomy, pollination, and natural history. Zootaxa. 4352: 1–160.

**Hadley, A. 2010**. CombineZP: Image stacking software.

**Harmon-Threatt, A. N., and S. D. Hendrix**. **2015**. Prairie restorations and bees: The potential ability of seed mixes to foster native bee communities. Basic and Applied Ecology. 16: 64–72.

**Hurd, P. D. 1979**. Superfamily Apoidea, pp. 1741–2209. *In* Krombein, K. V., Hurd, P.D., Smith, D.R., Burks, B.D. (eds.), Catalog of Hymenoptera in America North of Mexico. Volume 2. Smithsonian Institution Press, Washington, D. C.

**Kearns, C. A., and D. W. Inouye**. **1993**. Techniques for Pollination Biologists. University Press of Colorado, Niwot, CO.

**LaBerge, W. E. 1986**. The zoogeography of *Andrena* Fabricius (Hymenoptera: Andrenidae) of the Western Hemisphere, pp. 110–115. *In* Clamby, G.K., Pemble, R.H. (eds.), Prairie Past, Present. Futur. Proc. Ninth North Am. Prairie Conf. Tri-College University for Environmental Studies, Fargo, North Dakota.

**Lane, I. G., C. R. Herron-Sweet, Z. M. Portman, and D. P. Cariveau. 2020.** Floral resource diversity drives bee community diversity in prairie restorations along an agricultural landscape gradient. Journal of Applied Ecology. Available from https://doi.org/10.1111/1365-2664.13694.

**Lindsey, A. H. 1984**. Reproductive biology of Apiaceae. I. Floral visitors to *Thaspium* and *Zizia* and thier importance in pollination. American Journal of Botany. 71: 375–387.

**Lindsey, A. H., and C. R. Bell**. **1985**. Reproductive biology of Apiaceae. II. Cryptic specialization and floral evolution in *Thaspium* and *Zizia*. American Journal of Botany. 72: 231–247.

**Michener, C. D. 2007**. The Bees of the World, 2nd ed. Johns Hopkins University Press, Baltimore.

**Mitchell, T. B. 1960**. Bees of the Eastern United States. Volume 1., North Carolina Experiment Station Technical Bulletin 141: 1– 538.

**Normandin, É., N. J. Vereecken, C. M. Buddle, and V. Fournier**. **2017**. Taxonomic and functional trait diversity of wild bees in different urban settings. PeerJ. 5: e3051.

**Pennarola, P. 2019**. The revery alone won't do: Fire grazing, and other drivers of bee communities in remnant tallgrass prairie. M.S. thesis, University of Minnesota.

**Ribble, D. W. 1968**. Revisions of two subgenera of *Andrena*: *Micrandrena* Ashmead and *Derandrena*, new subgenus (Hymenoptera: Apoidea). Bulletin of the University of Nebraska State Museum. 8: 237–394.

**Ritchie, A. D., R. Ruppel, and S. Jha**. **2016**. Generalist behavior describes pollen foraging for perceived oligolectic and polylectic bees. Environmental Entomology. 45: 909–919.

**Robertson, C. 1891**. Descriptions of new species of North American bees. Transactions of the American Entomological Society. 18: 49–66.

**Robertson, C. 1895**. Notes on bees, with descriptions of new species.—Third paper. Transac-

tions of the American Entomological Society. 22: 115–128.

**Robertson, C. 1897**. North American bees—Descriptions and synonyms. Transactions of the Academy of Science of St. Louis. 7: 315–356.

**Robertson, C. 1902**. Synopsis of Andreninæ. Transactions of the American Entomological Society. 28: 187–194.

**Robertson, C. 1905**. Some new or little-known bees.—V. Canadian Entomologist. 37: 236–237.

**Robertson, C. 1926**. Revised list of oligolectic bees. Ecology. 7: 378–380.

**Russell, K. N., H. Ikerd, and S. Droege**. **2005**. The potential conservation value of unmowed powerline strips for native bees. Biological Conservation. 124: 133–148.

**Smith, B. A., R. L. Brown, W. Laberge, and T. Griswold**. **2012**. A faunistic survey of bees (Hymenoptera: Apoidea) in the Black Belt Prairie of Mississippi. Journal of the Kansas Entomological Society. 85: 32–47.

**Viereck, H. L. 1916**. The Hymenoptera, or wasp-like insects, of Connecticut. Connecticut State Geological and Natural History Survey Bulletin. 22: 1–824.

**Wolf, A. T., and J. S. Ascher**. **2008**. Bees of Wisconsin (Hymenoptera: Apoidea: Anthophila). Great Lakes Entomologist. 41: 129–168.

**Wood, T. J., and S. P. M. Roberts**. **2018**. Constrained patterns of pollen use in Nearctic *Andrena* (Hymenoptera: Andrenidae) compared with their Palaearctic counterparts. Biological Journal of the Linnean Society. 124: 732–746.

# First Records of *Megachile apicalis* (Hymenoptera: Megachilidae) for Illinois Found in Heavily Urbanized Areas Within the City of Chicago

Andrea M. Gruver[1, 2, *], & Paul J. CaraDonna[1,2]

[1] Negaunee Institute for Plant Conservation Science and Action, Chicago Botanic Garden, Glencoe, IL, USA 60022
[2] Plant Biology & Conservation, Northwestern University, Evanston, IL 60208
[*] Corresponding author: (e-mail: andreagruver@gmail.com)

**Abstract**

We provide the first record of the non-native *Megachile apicalis* Spinola (Hymenoptera: Megachilidae) in Illinois. Thirty *M. apicalis* specimens were collected in the summer of 2018 in a highly urban area of Chicago, IL, USA. Our findings suggest that the range of *M. apicalis* appears to be rapidly expanding across North America, inward from both the East and Western Coasts. *Megachile apicalis* in Chicago has a broad activity period, can take advantage of abundant non-native floral food resources and has competitive nesting behavior, all of which may facilitate its successful establishment in disturbed urban environments, as well as its continued spread across North America.

**Keywords:** new record, *Megachile*, invasive species, bees, urban ecology, Chicago

---

Non-native bee species continue to spread globally. Over 83 bee species have established outside of their native ranges worldwide and at least 40 species have established in North America alone (Cane 2003, Russo 2016, Gibbs and Dathe 2017, Martins et al. 2017). Non-native species can have detrimental effects on native ecosystems, yet the consequences of non-native bees are generally unknown, especially for unintentionally introduced species (Goulson 2003, Russo 2016). Some non-native bee species may be well-equipped to navigate harsh, urban landscapes, with a number of non-native species recorded exclusively in urban environments (Russo 2016, Portman et al. 2019). As urbanization increases worldwide, non-native species may be able to exploit urban environments compounding threats to local biodiversity (McKinney 2008, 2006).

Bees in the genus *Megachile* represent over 1,500 described species in 56 subgenera, accounting for 1/3 of all bees in the family Megachilidae (Michener 2007, Ascher and Pickering 2020). At least thirteen *Megachile* species are considered non-native across the globe, having expanded outside their native range—the most of any genus of bees (Russo 2016). There are at least six non-native *Megachile* species in the continental United States, many of which appear to be expanding rapidly, including *Megachile apicalis* Spinola (Hymenoptera: Megachilidae). *Megachile apicalis* is a cavity nesting species native to Europe, North Africa, and the Middle East and was accidently introduced to North America. The first record of *M. apicalis* in North America is from two specimens from Virginia, one from 1931 and the other from 1883, although the latter specimen is somewhat questionable. Following the first collections, *M. apicalis* was recorded in California in 1982 (Mitchell 1962, Cooper 1984) and expanded throughout parts of the west (Sheffield et al. 2011b, Droege 2015). *Megachile apicalis* has since been reported in Pennsylvania in 1996 (Donovall and VanEngelsdorp 2010), Ohio in 2010 (Sivakoff et al. 2018), Michigan in 2013 (Gibbs et al. 2017) and Missouri (date unknown; Camilo et al. 2017). The spread of *M. apicalis* across North America has been attributed, in part, to being transported in managed *Megachile rotundata* (Fabricius) pollinator nesting tubes (a close relative, which was accidently introduced in the 1940's, and is now widely managed as an important alfalfa pollinator; Pitts-Singer and Cane 2011, Droege 2015, Russo 2016).

Here, we report the first records of the non-native leafcutter bee, *M. apicalis* in Illinois, which were found in a heavily urbanized area within the City of Chicago. We provide detailed information on the urban ecosystem within which they were recorded, describe several ecologically relevant traits for *M. apicalis*, and discuss the potential effects *M. apicalis* may have on existing bee communities.

## Materials and Methods

*Megachile apicalis* specimens were collected during the 2018 growing season

as part of a larger study investigating bee communities along an urbanization gradient in the Chicago Metropolitan Area (Gruver and CaraDonna *unpublished*). Sampling occurred at eight different sites along an urbanization gradient that followed the Union Pacific North Metra Train Line. We quantified urbanization as the percent impervious surface within a 500 m radius from the sampling location. Across the eight study sites, impervious surface ranged from 69% within the urban core of Chicago to 15% in suburban areas outside of the city. Floral resources were sampled at each site four times from June to August 2018. To quantify the availability of floral resources, we divided each site into eight 25 × 3m sections; within each section, we recorded the identities of flowering species and counted the number of flowering inflorescences of each species. In addition, we recorded the percent cover of all flowering plants, non-flowering vegetation, bare ground, impervious surface, and lawn in each section.

Bees were sampled four times throughout the 2018 season (June–August) at each site using hand nets and pan trapping. *Megachile apicalis* specimens were identified using discover life keys, the Bees of Eastern United States, and the *Megachile* of Canada and Alaska (Mitchell 1962, Sheffield et al. 2011a, Andrus and Droege 2020). Bee specimens from this project are permanently deposited within the Arthropod Collection at the Field Museum (Chicago, IL, USA). Photographs of *M. apicalis* specimens were taken using a Canon EOS rebel T6i camera mounted on a Zeiss stemi 2000c stereomicroscope. Multiple photos were taken of specimens and stacked to create a composite image using Adobe Photoshop 2018 software (Adobe Systems Inc., San Jose, CA).

## Results

During the summer of 2018, we collected 30 *M. apicalis* specimens. All specimens were collected from a single site along the urbanization gradient: the Clybourn Metra Train Station (here after Clybourn), Chicago, Cook County, Illinois (41°55′1.2″N, 87°40′4.″W). Of all eight sites sampled across the urbanization gradient, Clybourn exhibits the greatest surrounding percent impervious surface (69%) and is closest to the city center (5.5 km from downtown Chicago). *Megachile apicalis* specimens were collected during all sampling periods across the summer of 2018 (28 June, 11 July, 27 July, and 10 August). Among the *M. apicalis* specimens collected, 47% (14) were female and 53% (16) male. Most specimens were collected in pan traps 77% (23), and 23% (7) collected while foraging on flowers. We recorded male *M. apicalis* foraging on four flowering plant species, all of which are non-native to North America. Three of these flowering plant species were members of the Fabaceae, *Medicago lupulina*, *Melilotus albu*s, and *Melilotus officinalis,* and one was a member of Asteraceae, *Centaurea stoebe*. We did not record any females foraging on plants—all were caught in bee bowls. The Clybourn site was dominated by non-native plant species (87%) over the growing season, the most of any of the sampled sites in 2018. The non-native flowering plants were also more abundant than native flowering plants, and made up 92.5% of the inflorescences over the course of the season at the Clybourn site.

In addition to *M. apicalis*, we collected several other adventive *Megachile* species across our urbanization gradient: *Megachile rotundata*, *Megachile sculpturalis* Smith, and *Megachile pusilla* Pérez. Apart from those in the genus *Megachile*, we also recorded the non-native *Pseudoanthidium nanum* (Mocsáry), *Hylaeus punctatus* (Brullé), *Hylaeus leptocephalus* (Morawitz), *Hylaeus hyalinatus* Smith, *Chelostoma rapunculi* (Lepeletier), *Anthidium manicatum* (L.), *Anthidium oblongatum* (Illiger), *Andrena wilkella* (Kirby) and *Apis mellifera* L.

***Species description.*** Members of the genus *Megachile* represent a group of robust bees that can be separated from other genera by the combination of the following characteristics: a lack of maculations on the integument, arolia absent, and abdominal tergum 1 (T1) with an anterior face creating a concave appearance (Mitchell 1962). Detailed descriptions of *M. apicalis* are provided in Mitchell (1962) and Sheffield et al. (2011a). Females of *M. apicalis* (Fig. 1) can be identified from other North American *Megachile* by the following combination of distinct characteristics: (i) clypeus with a distinct thick projection medially (Fig. 1C); (ii) abdominal terga 2 and 3 (T2 and T3) with lateral oval impressions that are shallow and impunctate (Fig. 1D); (iii) abdominal sternum 4 (S4) with a few black hairs and sterna 5 and 6 (S5 and S6) with hairs entirely black (Fig. 1E) (Mitchell 1962, Parker 1978, Sheffield et al. 2011a). The average intertegular distance for female *M. apicalis* specimens collected in Chicago was 2.4mm (± 0.18).

Males of *M. apicalis* (Fig. 2) can be more difficult to distinguish. However, male *M. apicalis* can be readily identified by the combination of the following two characteristics: (i) T2 and T3 with lateral oval impressions (Fig. 2C), and (ii) T5 with black upright hairs (Fig. 2D) (Mitchell 1962, Parker 1978, Sheffield et al. 2011a). The average intertegular distance for male *M.*

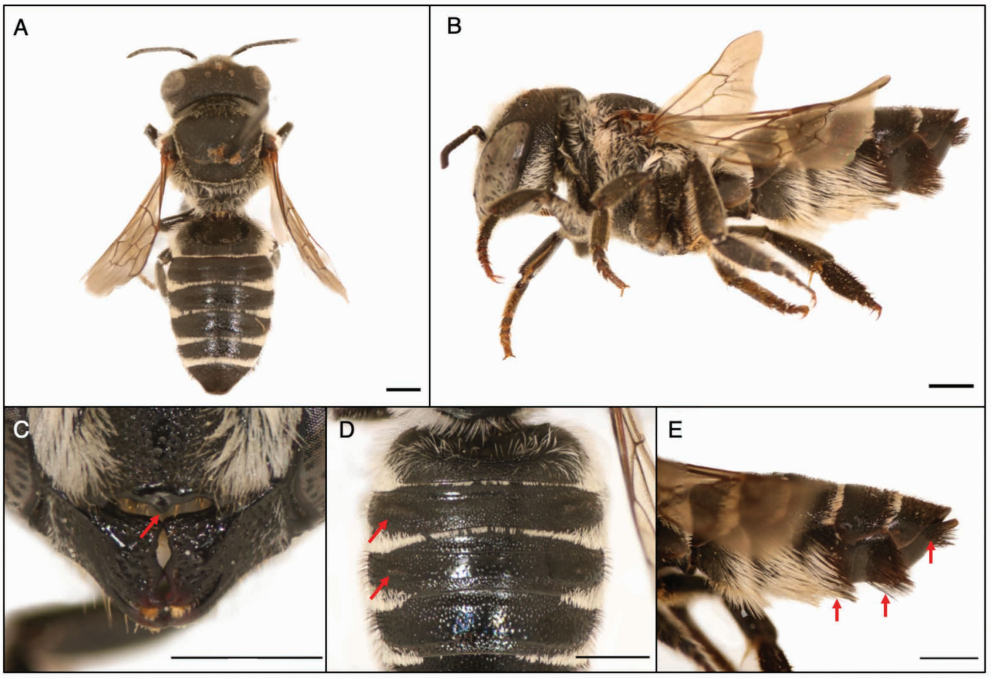


Figure 1: Female *Megachile apicalis*: A) dorsal view B) lateral view C) clypeus with medial projection D) dorsal view of abdomen showing terga 2 and 3 with shallow oval impressions E) lateral view of abdomen showing sterna 4, 5, and 6 with dark hairs. Scale bars = 1mm.

*apicalis* specimens collected in Chicago was 2.1mm (± 0.19).

## Discussion

These first records of *M. apicalis* in Illinois found within the City of Chicago suggest that this non-native bee is fully capable of taking advantage of extreme, urban environments. Across our urbanization gradient, with sites ranging from 15–69% impervious surface, *M. apicalis* was found exclusively at the site exhibiting the most extreme levels of urbanization (Clybourn). We observed *M. apicalis* across all four of our sampling periods from late June until early August, indicating that this bee has a relatively broad flight activity period. We also observed *M. apicalis* foraging only from non-native flowering plant species, which were abundant at this urban sites.

One of the plants on which we observed *M. apicalis* foraging was *Centaurea stoebe* (Asteraceae). *Megachile apicalis* has been shown to have strong preferences for plants in the Cynareae tribe, including *C. stoebe* (Müller and Bansac 2004). Interestingly, *M. apicalis* has also been documented along rail lines in Michigan foraging on *C. stoebe* and it has been suggested that this non-native plant may facilitate the spread of *M. apicalis* (Gibbs et al. 2017). Furthermore, *C. stoebe* was only documented at the Clybourn site, which may partly explain why this was the only site in which *M. apicalis* was observed. The apparent preference of *M. apicalis* for non-native plant species suggests it may be able to flourish in highly disturbed areas with high abundance of non-native floral resources. In addition to the availability of floral resources in highly urban areas, other studies strongly suggest that urban areas likely contain numerous nesting opportunities for cavity nesting bees, including non-native species like *M. apicalis* (Matteson et al. 2008, Fortel et al. 2014).

*Megachile apicalis* may be well suited to exploit the urban environment given its behavior and ecological traits, potentially impacting other native bee species. Although we do not know the extent of the impact of non-native bee species, there is evidence that non-native species in the genus *Megachile* may compete strongly with native bees for nesting resources. Bees in the subgenus *Eutricharaea* (a non-native subgenus in North America), including *M. apicalis* and *M. rotundata*, create nests in cavities with

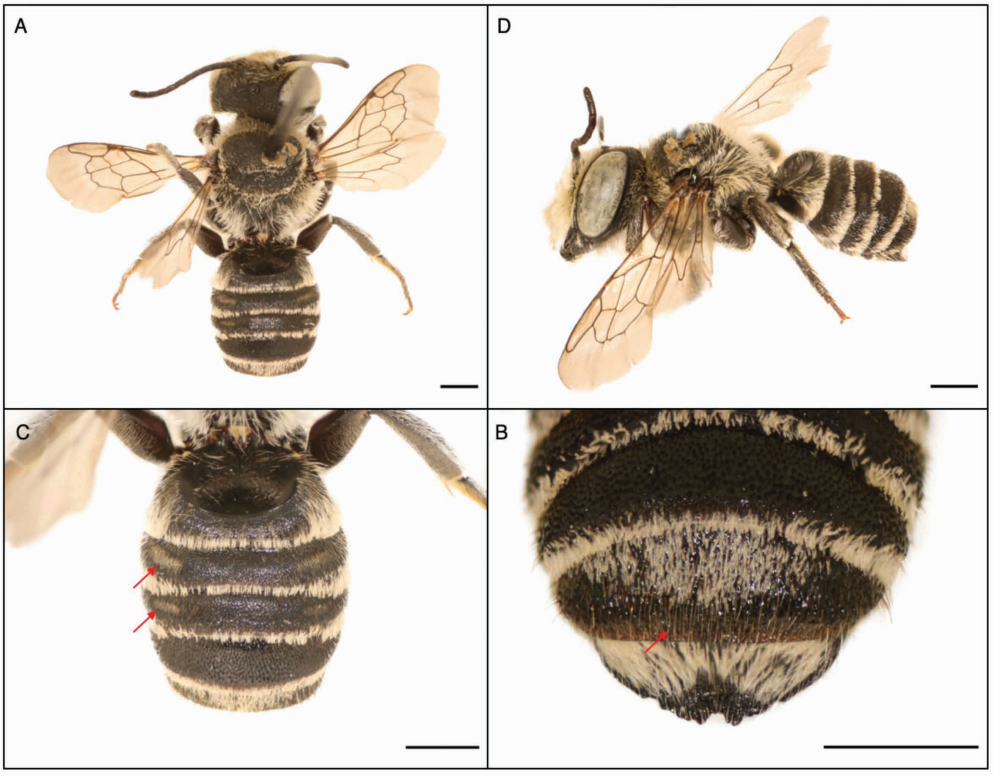


Figure 2: Male *Megachile apicalis*: A) dorsal view B) lateral view C) dorsal view of abdomen showing terga 2 and 3 with shallow oval impressions D) dorsal view of abdominal terga 5 with dark upright hairs. Scale bars = 1mm.

leaf disks that are glued together that act to protect brood from nest parasites and allow them to nest in a variety of cavities (Trostle and Torchio 1994, Frankie et al. 1998). This nesting flexibility and added protection may give them a competitive advantage against other *Megachile* species (Barthell et al. 1998, Frankie et al. 1998). In addition to nesting structure, *M. apicalis* females have been documented displaying aggressive nest usurpation behavior towards other bees, which may increase its ability to acquire and maintain nests (Barthell and Thorp 1995). *Megachile apicalis* has been documented competing with *M. rotundata* for nest sites, but it is not clear the extent to which *M. apicalis* may compete with locally native *Megachile* species that may have different nesting requirements than non-native *Megachile* species (Frankie et al. 1998). Taken together, *M. apicalis* in Chicago has a broad activity period, can take advantage of abundant non-native floral food resources, and has competitive nesting behavior, all of which may facilitate its successful establishment in disturbed urban environments, as well as its continued spread across North America.

Urban centers have been suggested to be refuges for bees (Hall et al. 2017). Although cities can potentially harbor a diversity of bee species, in some cases, cities appear to have larger proportion of non-native bees compared to less disturbed areas (Fetridge et al. 2008, Matteson et al. 2008, Fitch et al. 2019, Wilson and Jamieson 2019). In addition, some non-native bee species have only been documented in highly disturbed areas, as is the case here with *M. apicalis*, suggesting cities may be a favorable habitat for them (O'Brien et al. 2012, Portman et al. 2019). As urbanization continues to grow worldwide, it is important to monitor the spread of non-native bee species to help us determine the potential impacts they may pose on native bee species.

## Acknowledgments

We thank John Ascher for confirming our initial identification of *Megachile apicalis* on BugGuide. Thanks to Maya Dutta,

Elaine Jiang, and Alicia Wala for fieldwork assistance. We also thank the Iler + Cara-Donna Lab Group and two anonymous reviewers for constructive comments on the manuscript. Research was supported by an NSF Graduate Research Fellowship (DGE-1842165 to A.M.G.) and the Northwestern University Plant Biology and Conservation Research Award (to A.M.G.).

## Literature cited

**Andrus, R., and S. Droege. 2020.** Guide to the Female *Megachile* of Eastern North America. Available from https://www.discoverlife.org/mp/20q?guide=*Megachile*_female.

**Ascher, J. S., and J. Pickering. 2020.** Discover Life bee species guide and world checklist (Hymenoptera: Apoidea: Anthophila). Available from https://www.discoverlife.org/mp/20q?search=Apoidea.

**Barthell, J. F., G. W. Frankie, and R. W. Thorp. 1998.** Invader Effects in a Community of Cavity Nesting Megachilid Bees (Hymenoptera: Megachilidae). Environmental Entomology 27: 240–247.

**Barthell, J., and R. Thorp. 1995.** Nest Usurpation Among Females of an Introduced Leaf-Cutter Bee, *Megachile apicalis*. Southwestern Entomologist 20: 117–124.

**Camilo, G. R., P. A. Muñiz, M. S. Arduser, and E. M. Spevak. 2017.** A Checklist of the Bees (Hymenoptera: Apoidea) of St. Louis, Missouri, USA. Journal of the Kansas Entomological Society 90: 175–188.

**Cane, J. H. 2003.** Exotic non-social bees (Hymenoptera: Apoidea) in North America: ecological implications. In *For non-native crops, whence pollinators of the future?*., ed. K. Strickler and J. H. Cane, pp. 113–126. Lanham, MD: Entomological Society of America.

**Cooper, K. W. 1984.** Discovery of First Resident Population of the European Bee, *Megachile apicalis* in the United States (Hymenoptera: Megachilidae). Entomological News 95: 225–226.

**Donovall, L. R., and D. vanEngelsdorp. 2010.** A Checklist of the Bees (Hymenoptera: Apoidea) of Pennsylvania. Journal of the Kansas Entomological Society 83: 7–24.

**Droege, S. 2015.** The Very Handy Manual How to Catch and Identify Bees and Manage a Collection. Available from https://bee-health.extension.org/wp-content/uploads/2019/08/TheVeryHandyBeeManual.pdf

**Fetridge, E. D., J. S. Ascher, and G. A. Langellotto. 2008.** The Bee Fauna of Residential Gardens in a Suburb of New York City (Hymenoptera: Apoidea). Annals of the Entomological Society of America 101: 1067–1077.

**Fitch, G., C. J. Wilson, P. Glaum, C. Vaidya, M.-C. Simao, and M. A. Jamieson. 2019.** Does urbanization favour exotic bee species? Implications for the conservation of native bees in cities. Biology Letters 15:20190574. Available from https://doi.org/10.1098/rsbl.2019.0574

**Fortel, L., M. Henry, L. Guilbaud, A. L. Guirao, M. Kuhlmann, H. Mouret, O. Rollin, and B. E. Vaissière. 2014.** Decreasing Abundance, Increasing Diversity and Changing Structure of the Wild Bee Community (Hymenoptera: Anthophila) along an Urbanization Gradient. PLoS ONE 9:e104679.

**Frankie, G. W., R. W. Thorp, L. E. Newstrom-Lloyd, M. A. Rizzardi, J. F. Barthell, T. L. Griswold, J.-Y. Kim, and S. Kappagoda. 1998.** Monitoring Solitary Bees in Modified Wildland Habitats: Implications for Bee Ecology and Conservation. Environmental Entomology 27: 1137–1148.

**Gibbs, J., J. S. Ascher, M. G. Rightmyer, and R. Isaacs. 2017.** The bees of Michigan (Hymenoptera: Apoidea: Anthophila), with notes on distribution, taxonomy, pollination, and natural history. Zootaxa 4352: 1–160.

**Gibbs, J., and H. H. Dathe. 2017.** First records of *Hylaeus* ( *Paraprosopis* ) *pictipes* Nylander, 1852 (Hymenoptera: Colletidae) in North America. Check List 13:2116. Available from https://www.biotaxa.org/cl/article/view/13.3.2116

**Goulson, D. 2003.** Effects of Introduced Bees on Native Ecosystems. Annual Review of Ecology, Evolution, and Systematics 34: 1–26.

**Hall, D. M., G. R. Camilo, R. K. Tonietto, J. Ollerton, K. Ahrné, M. Arduser, J. S. Ascher, K. C. R. Baldock, R. Fowler, G. Frankie, D. Goulson, B. Gunnarsson, M. E. Hanley, J. I. Jackson, G. Langellotto, D. Lowenstein, E. S. Minor, S. M. Philpott, S. G. Potts, M. H. Sirohi, E. M. Spevak, G. N. Stone, and C. G. Threlfall. 2017.** The city as a refuge for insect pollinators: Insect Pollinators. Conservation Biology 31: 24–29.

**Martins, K. T., A. Gonzalez, and M. J. Lechowicz. 2017.** Patterns of pollinator turnover and increasing diversity associated with urban habitats. Urban Ecosystems 20: 1359–1371.

**Matteson, K. C., J. S. Ascher, and G. A. Langellotto. 2008.** Bee Richness and Abundance in New York City Urban Gardens. Annals of the Entomological Society of America 101: 140–150.

**McKinney, M. L. 2006.** Urbanization as a major cause of biotic homogenization. Biological Conservation 127: 247–260.

**McKinney, M. L. 2008.** Effects of urbanization on species richness: A review of plants and animals. Urban Ecosystems 11: 161–176.

**Michener, C. 2007.** The Bees of the World. 2nd edition. John Hopkins University Press, Baltimore and London.

**Mitchell, T. B. 1962.** Bees of the Eastern United States. Volume 2., North Carolina Experimental Station Technical Bulletin 152: 1–557.

**Müller, A., and N. Bansac. 2004.** A specialized pollen-harvesting device in western palaearctic bees of the genus *Megachile* (Hymenoptera, Apoidea, Megachilidae). Apidologie 35: 329–337.

**O'Brien, M. F., D. R. Swanson, and J. Monsma. 2012.** *Anthidium Oblongatum* (Apoidea: Megachilidae) Confirmed as a Michigan Resident, with Notes on Other Michigan *Anthidium* Species. The Great Lakes Entomologist 45: 102–105.

**Parker, F. D. 1978.** An illustrated key to alfalfa leafcutter bees *Eutricharaea* (Hymenoptera: Megachilidae). Pan-Pacific Entomologist 54: 61–64.

**Pitts-Singer, T. L., and J. H. Cane. 2011.** The Alfalfa Leafcutting Bee, *Megachile rotundata:* The World's Most Intensively Managed Solitary Bee. Annual Review of Entomology 56: 221–237.

**Portman, Z. M., S. J. Burrows, T. Griswold, M. Arduser, A. J. Irber, R. K. Tonietto, and D. P. Cariveau. 2019.** First Records of the Adventive *Pseudoanthidium nanum* (Mocsáry)(Hymenoptera: Megachilidae) in Illinois and Minnesota, with Notes on its Identification and Taxonomy. The Great Lakes Entomologist 52: 12–29.

**Russo, L. 2016.** Positive and Negative Impacts of Non-Native Bee Species around the World. Insects 7:69. Available from https://doi.org/10.3390/insects7040069

**Sheffield, C., C. Ratti, L. Packer, and T. Griswold. 2011a.** Leafcutter and mason bees of the genus *Megachile* Latreille (Hymenoptera: Megachilidae) in Canada and Alaska. Canadian Journal of Arthropod Identification 18: 1–106.

**Sheffield, C. S., S. Dumesh, and M. Cheryomina. 2011b.** *Hylaeus punctatus* (Hymenoptera: Colletidae), a bee species new to Canada, with notes on other non-native species. The Journal of Entomological Society of Ontario 142: 29–43.

**Sivakoff, F. S., S. P. Prajzner, and M. M. Gardiner. 2018.** Unique Bee Communities within Vacant Lots and Urban Farms Result from Variation in Surrounding Urbanization Intensity. Sustainability 10:1926. Available from https://doi.org/10.3390/su10061926

**Trostle, G., and P. F. Torchio. 1994.** Comparative Nesting Behavior and Immature Development of *Megachile rotundata* (Fabricius) and *Megachile apicalis* Spinola (Hymenoptera: Megachilidae). Journal of the Kansas Entomological Society 67: 53–72.

**Wilson, C. J., and M. A. Jamieson. 2019.** The effects of urbanization on bee communities depends on floral resource availability and bee functional traits. PLOS ONE 14:e0225852.

# New State Species Records and Noteworthy Re-captures of Michigan (USA) Trichoptera

David C. Houghton
Department of Biology, Hillsdale College, 33 East College Street, Hillsdale, MI 49242
(e-mail: david.houghton@hillsdale.edu)

**Abstract**

Ten caddisfly species are reported from Michigan for the first time, including 5 in the family Limnephilidae [*Limnephilus femoralis* Kirby, *L. thorus* Ross, *Pycnopsyche aglona* Ross, *P. circularis* (Provancher), and *P. limbata* (McLachlan)], 2 in the Polycentropodidae (*Cernotina pallida* (Banks) and *Holocentropus glacialis* Ross), 1 in the Hydroptilidae (*Hydroptila fiskei* Blickle), 1 in the Leptoceridae (*Triaenodes perna* Ross), and 1 in the Rhyacophilidae (*Rhyacophila angelita* Banks). An additional 5 unique species, including several not seen in MI for over 70 years, are confirmed to be extant in the state, most notably *Plectrocnemia sabulosa* (Leonard & Leonard), MI's only known endemic caddisfly. The state is now known to contain 305 caddisfly species.

**Keywords:** Trichoptera, caddisfly, Michigan, species, record

---

The caddisflies of Michigan are relatively well known. The most recent statewide checklist (Houghton et al. 2018) documented 295 species, including 41 state species records. These species were compiled from almost 700 collections dating back to the 1930s, and including substantial sampling during the 2010s.

The vast majority of the aforementioned collections have occurred during the summer months. For example, my students and I have sampled several hundred lakes and streams throughout the state since 2010; however, due to inclement weather and the beginning of the academic year, only 6 of these sites have been sampled during the autumn flight period. While the autumn flight period includes far fewer species than the summer flight period, it also features many species unique to it, particularly within the families Limnephilidae and Thremmatidae (Houghton 2018).

Furthermore, MI is a large state that still contains regions with low collecting effort. For example, when I began this study only 20 caddisfly species had been reported from the remote Huron Mountains of the Upper Peninsula (Woods 2015). Thus, the objective of this study was to visit areas and seasons not normally collected, to attempt to find additional species not documented by the recent state checklist.

## Materials and Methods

Most of the records noted herein are from sampling in the Huron Mountains. Other records have come from a variety of other collecting sites throughout the state (Fig. 1). In particular, a series of September collections was made at sites in the Huron Mountains and Munising area of the Upper Peninsula, and the Manistee River region of the Lower Peninsula. Dates of these collections ranged 05–30 September 2019. All reported specimens were collected using 8-watt portable ultraviolet light traps. These traps were deployed at dusk and retrieved 1–2 h later. All reported specimens are housed in the Hillsdale College Insect Collection.

## Results

A total of 10 new species records for MI are reported herein. In addition, 5 other unique species, some not seen in MI for over 70 years, are confirmed to be extant in the state. Collecting information is included with each species account. The number assigned to each species corresponds to the collecting localities in Fig. 1.

### Family Hydropsychidae

**1. *Arctopsyche ladogensis* (Kolenati, 1859).** This species has a Holarctic distribution (Cushman 2014). It is known in MI from several collections of larvae in Houghton, Marquette, and Ontonagon Counties, mostly from the 1930s–1940s, and most recently by Yanoviak and McCafferty (1996). The only reported adult collection was from the North Branch of the Otter River in Houghton County in 1949. It is reported herein from

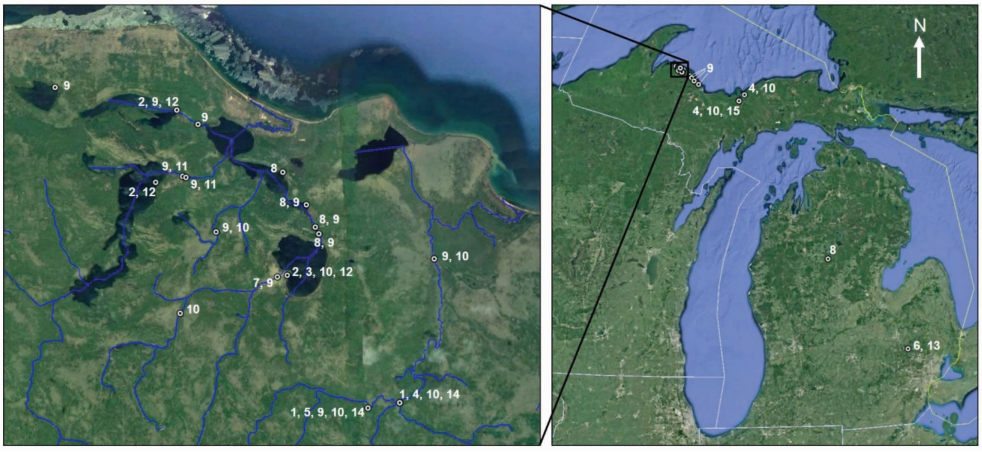


**Figure 1**. Maps of MI collecting localities for the species noted herein, showing detail of the Huron Mountains habitats. Numbers correspond to the numbers before each species account; thus, the map shows how many species were found at each locality. Basemaps: © 2020 Google Image, © 2020 TerraMetrics Image NOAA.

two adjacent sites of the Salmon Trout River in Marquette County. Only female specimens were collected, which were identified using Givens and Ruiter (2015). Interestingly, specimens were abundant in late June 2019, but absent in July of the same year. The species has not been recorded from any adjacent states or provinces, despite substantial searching in expected habitats of northern MN and WI (Schmude and Hilsenhoff 1986, MacLean 1995, Houghton 2012).

MATERIAL EXAMINED. Marquette County: Salmon Trout River, below Middle Rapids, Huron Mountain Club, 46.8100, –87.8245, 26 June 2019, Houghton et al., uv light, 5♀; Salmon Trout River, Lower Dam, Huron Mountain Club, 46.8114, –87.8125, 26 June 2019, 79 ♀.

### Family Hydroptilidae

**2. *Hydroptila fiskei* Blickle 1963.** NEW STATE RECORD. This species is reported herein from MI for the first time based on specimens collected from 4 lakes and 1 stream in Marquette County. It has previously been recorded from the eastern US (Rasmussen and Morse 2018), as well as northern MN and southern ON (unpublished data).

MATERIAL EXAMINED Marquette County: Ives Lake, Ives Lake Road, Huron Mountain Club, at Stonehouse, 46.8439, –87.8547, 23 July 2019, Houghton et al., uv light, 3♂; Rush Lake, East side boathouse, Huron Mountain Club, 46.8869, –87.8967, 29 June 2019, Houghton et al., uv light, 2♂; Mountain Lake, boathouse on Mountain Lake Road, Huron Mountain Club, 46.8681, –87.9043, 24 July 2019, Houghton et al., uv light, 2♂; Second Pine Lake, east boathouse, Huron Mountain Club, 46.8705, –87.8567, 19 September 2019, Houghton et al., uv light, 1♂.

### Family Leptoceridae

**3. *Triaenodes perna* Ross 1938.** NEW STATE RECORD. This species in known throughout the eastern US and southern Canada, including IL, OH, and ON (Rasmussen and Morse 2018). It is reported herein from MI for the first time based on a single specimen collected from Ives Lake in Marquette County.

MATERIAL EXAMINED. Marquette County: Ives Lake, Ives Lake Road, at Stonehouse, Huron Mountain Club, 46.8439, –87.8547, 23 July 2019, Houghton et al., uv light, 1♂.

### Family Limnephilidae

**4. *Asynarchus rossi* (Leonard and Leonard 1949).** This species was first described from specimens collected from the Au Sable River in Crawford County. Leonard and Leonard (1949) also reported specimens from Hunt Creek in Montmorency County. It is reported herein from several collections in Alger and Marquette Counties. These records constitute the first MI collections of the species in over 70 years, as well as the first from the Upper Peninsula. The species is known primarily from the northcentral US, including MN and WI (Rasmussen and Morse 2018). All previous adult collections have occurred during autumn (Karl and Hilsenhoff 1979, Houghton 2012).

MATERIAL EXAMINED. Alger County: Wagner Falls of Wagner Creek, MI 94/28, public ac-

cess, 46.3881, –86.6461, 21 September 2019, Houghton et al., uv light, 4♂; Alger Falls, MI 94, 46.3933, –86.6482, 21 September 2019, Houghton et al., uv light, 19♂, 4♀; Marquette County: Salmon Trout River, Lower Dam, Huron Mountain Club, 46.8114, –87.8125, 16 September 2019, Houghton et al., uv light, 5♂.

**5. *Limnephilus femoralis* Kirby 1837.** NEW STATE RECORD. This species has a northern Holarctic distribution, and has previously been recorded from ME and WA (Rasmussen and Morse 2018), as well as WI (unpublished data). It is reported herein from MI for the first time based on a single specimen collected from the Salmon Trout River in Marquette County.

MATERIAL EXAMINED. Marquette County: Salmon Trout River, Middle Falls, Huron Mountain Club, 46.8100, –87.8245, 28 June 2015, Houghton et al., uv light, 1♀.

**6. *Limnephilus perpusillus* Walker 1852**. This species is known from MI based on a series of collections from the ES George Reserve in Livingston County during the 1940s and maintained in the University of Michigan Museum of Zoology. It is reported herein based on a single specimen from Duck Creek in Oakland County. This record constitutes the first MI collection of the species in over 70 years. The species is known from throughout Canada and the northern US, including MN, OH, ON, and WI (Rasmussen and Morse 2018).

MATERIAL EXAMINED. Oakland County: Duck Creek, Bird Road, near Ortonville, 42.8125, –83.4706, 06 July 2015, Houghton et al., uv light, 1♂.

**7. *Limnephilus thorus* Ross 1938.** NEW STATE RECORD. This species is reported herein from MI collected from Elm Creek in Marquette County. While known primarily from the western US, it has also been reported from MN (Rasmussen and Morse 2018). All collections from MI and MN have occurred during autumn (Houghton 2012).

MATERIAL EXAMINED. Marquette County: Elm Creek, near Stonehouse at Ives Lake, Huron Mountain Club, 46.8439, –87.8586, 18 September 2019, Houghton et al., uv light, 1♂.

**8. *Pycnopsyche aglona* Ross 1941**. NEW STATE RECORD. This species is known from southern Canada and the northeastern US, including MN, OH, ON, and WI (Rasmussen and Morse 2018). It is reported herein from MI for the first time based on collections in Clare and Marquette Counties. All collections occurred in autumn.

MATERIAL EXAMINED. Clare County: Cranberry Creek, West Wilson Road / Muskegon Road, 44.1326, –84.9474, 05 September 2019, Houghton et al., uv light 23♂, 2♀; Marquette County: River Styx, base of cascade, Huron Mountain Club, 46.8550, –87.8428, 16 September 2019, Houghton et al., uv light, 6♂; River Styx, access bridge off of main road, Huron Mountain Club, 46.8567, –87.8446, 16 September 2019, Houghton et al., uv light, 79♂ 6♀; Third Pine Lake, eastern shore picnic area, Huron Mountain Club, 46.8626, –87.8475, 19 September 2019, Houghton et al., uv light, 7♂; Second Pine Lake, east boathouse, Huron Mountain Club, 46.8705, –87.8567, 19 September 2019, Houghton et al., uv light, 1♂.

**9. *Pycnopsyche circularis* (Provancher 1877).** NEW STATE RECORD. This species is known from the eastern US, including OH and WI (Rasmussen and Morse 2018). It is reported herein from MI for the first time based on many collections throughout Marquette County. All collections occurred in autumn.

MATERIAL EXAMINED. Marquette County: River Styx, base of cascade, Huron Mountain Club, 46.8550, –87.8428, 16 September 2019, Houghton et al., uv light, 16♂; Salmon Trout River, entrance Bridge, Huron Mountain Club, 46.8485, –87.7989, 16 September 2019, Houghton et al., uv light, 21♂; Salmon Trout River, Middle Falls, Huron Mountain Club, 46.8100, –87.8245, 16 September 2019, Houghton et al., uv light, 13♂; River Styx, access bridge off of main road, Huron Mountain Club, 46.8567, –87.8446, 16 September 2019, Houghton et al., uv light, 23♂; Howe Lake, Boathouse on northeast corner, Huron Mountain Club, 46.8932, –87.9436, 17 September 2019, Houghton et al., uv light, 1♂; Rush Lake, Boathouse on east side, Huron Mountain Club, 46.8869, –87.8967, 17 September 2019, Houghton et al., uv light, 27♂; Mountain Stream, at Bridge, Mountain Lake Road, Huron Mountain Club, 46.8699, –87.8946, 17 September 2019, Houghton et al., uv light, 18♂; Elm Creek, near Stonehouse at Ives Lake, Huron Mountain Club, 46.8439, –87.8586, 18 September 2019, Houghton et al., uv light, 13♂; Fisher Creek, off of Ives Lake Road, Huron Mountain Club, 46.8555, –87.8819, 18 September 2019, Houghton et al., uv light, 3♂; Rush Creek, Mountain Lake Road, Huron Mountain Club, 46.88361, –87.8889, 19 September 2019, Houghton et al., uv light, 6♂; Mountain Stream, below waterfall, off of Mountain Lake Road, Huron Mountain Club, 46.8692, –87.8933, 19 September 2019, Houghton et al., uv light, 6♂; Third Pine Lake, eastern shore picnic area, Huron Mountain Club, 46.8626, –87.8475, 19 September 2019, Houghton et al., uv light, 2♂; Little Garlic River, County Road 550 / Big Bay Road, 46.6739, –87.5412, 20 September 2019, Houghton et al., uv light, 5♂; Yellow Dog River, County Road 550, 46.7568, –87.6616, 20 September 2019, Houghton et al., uv light, 3♂; Big Garlic

River, dirt road off of County Road 550 / Big Bay Road, 46.6832, –87.5700, 20 September 2019, Houghton et al., uv light, 5♂; Wilson Creek, Highway 550 / Big Bay Road, 46.7288, –87.6216, 20 September 2019, Houghton et al., uv light, 1♂; Harlow Creek, County Road 550, 46.6327, –87.4753, 20 September 2019, Houghton et al., uv light, 1♂.

**10. *Pycnopsyche limbata* (McLachlan 1871).** NEW STATE RECORD. This species is known from southern Canada and the northeastern US, including MN, ON, and WI (Rasmussen and Morse 2018). It is reported herein from MI for the first time based on several collections in Alger and Marquette Counties. All collections occurred in autumn.

MATERIAL EXAMINED. Alger County: Wagner Falls, Wagner Creek, MI 94/28, public access, 46.3881, –86.6461, 21 September 2019, Houghton et al., uv light, 1♂; Falls, Miners River, 46.4747, –86.5314, 21 September 2019, Houghton et al., uv light, 4♂; Downstream of Miners Falls, near scenic overlook, 46.4748, –86.5314, 21 September 2019, Houghton et al., uv light, 7♂; Alger Falls, MI 94, 46.3933, –86.6482, 21 September 2019, Houghton et al., uv light, 2♂; Marquette County, Salmon Trout River, Lower Dam, Huron Mountain Club, 46.8114, –87.8125, 16 September 2019, Houghton et al., uv light, 1♂; Salmon Trout River, entrance Bridge, Huron Mountain Club, 46.8485, –87.7989, 16 September 2019, Houghton et al., uv light, 4♂; Salmon Trout River, Middle Falls, Huron Mountain Club, 46.8100, –87.8245, 16 September 2019, Houghton et al., uv light, 1♂; South Fork of Elm Creek, off of Ives Lake Road, Huron Mountain Club, 46.8345, –87.8955, 18 September 2019, Houghton et al., uv light, 2♂; Fisher Creek, off of Ives Lake Road, Huron Mountain Club, 46.8555, –87.8819, 18 September 2019, Houghton et al., uv light, 3♂; Ives Lake, Ives Lake Road, at Stonehouse, Huron Mountain Club, 46.8439, –87.8547, 19 September 2019, Houghton et al., uv light, 1♂.

### Family Odontoceridae

**11. *Psilotreta indecisa* (Walker, 1852).** This species was first reported from MI by Bright and Bidlack (1998) based on a series of adults from the Huron River in Oakland County, and from larval specimens from the Escanaba River in Delta County and the Little Siskiwit River in Keweenaw County. It is reported herein from 2 localities in close proximity to each other of Mountain Stream in Marquette County. Interestingly, specimens were abundant at both sites in late June 2019, but absent in July of the same year. The species is known from throughout Canada and the northern US, including MN, OH, ON, and WI (Rasmussen and Morse 2018).

MATERIAL EXAMINED. Marquette County: Mountain Stream, at Bridge on Mountain Lake Road, Huron Mountain Club, 46.8699, –87.8946, 25 June 2019, Houghton et al., uv light, 49♂; Marquette County: Mountain Stream, below waterfall on Mountain Lake Road, Huron Mountain Club, 46.8692, –87.8933, 25 June 2019, Houghton et al., uv light, 52♂, 2♀.

### Family Polycentropodidae

**12. *Cernotina pallida* (Banks 1904).** NEW STATE RECORD. This species is known primarily from the eastern coast of the US, but has also been found in OH and ON (Rasmussen and Morse 2018). It is reported herein from MI for the first time based on collections of three lakes in Marquette County.

MATERIAL EXAMINED. Marquette County: Ives Lake, Ives Lake Road, Huron Mountain Club, at Stonehouse, 46.8439, –87.8547, 23 July 2019, Houghton et al., uv light, 6♂; Rush Lake, East Boathouse, Huron Mountain Club, 46.8869, –87.8967, 29 June 2019, Houghton et al., uv light, 31♂; Mountain Lake, Boathouse on Mountain Lake Road, Huron Mountain Club, 46.8681, –87.9043, 24 July 2019, Houghton et al., uv light, 1♂.

**13. *Holocentropus glacialis* Ross 1938.** NEW STATE RECORD. This species was erroneously reported from MI by Leonard and Leonard (1949) based on a specimen of *Plectrocnemia cinerea* (Hagen) housed in the Illinois Natural History Survey that was misidentified as *H. glacialis* (Houghton et al. 2018). It is reported herein from MI for the first time based on two males collected from Duck Creek in Oakland County. It is also known from IL, IN, MN, and WI (Rasmussen and Morse 2018).

MATERIAL EXAMINED. Oakland County: Duck Creek, Bird Road, near Ortonville, 42.8125, –83.4706, 06 July 2015, Houghton et al., uv light, 2♂

**14. *Plectrocnemia sabulosa* (Leonard and Leonard 1949).** This species is MI's only known endemic caddisfly. It was described from specimens collected from the Au Sable River in Crawford County in 1948, found at several sites along Houghton Creek in Ogemaw County from 1951–1953 (Ellis 1962), and has not been reported anywhere since. It is reported herein from two adjacent sites of the Salmon Trout River in Marquette County. These records constitute the first captures of the species in almost 70 years, as well as the first from the Upper Peninsula. The latter observation

suggests that *P. sabulosa* may also exist in northern WI.

MATERIAL EXAMINED. Marquette County: Salmon Trout River, below Middle Rapids Huron Mountain Club, 46.8100, –87.8245, 26 June 2019, Houghton et al., uv light, 8♂; Salmon Trout River, Lower Dam, Huron Mountain Club, 46.8114, –87.8125, 26 June 2019 2♂.

### Family Rhyacophilidae

**15. *Rhyacophila angelita* Banks 1911**. NEW STATE RECORD. This species is found mostly in the western US and Canada, although it has also been reported from MN, NH, and QC (Rasmussen and Morse 2018). It is reported herein from MI for the first time based on a single specimen collected from Wagner Falls in Alger County.

MATERIAL EXAMINED. Alger County: Wagner Falls of Wagner Creek, MI 94/28, public access, 46.3881, –86.6461, 21 September 2019, Houghton et al., uv light, 1♂.

### Discussion

This paper demonstrates the importance of collecting throughout multiple flight seasons. For example, the 3 autumn-flying species of *Pycnopsyche* documented herein for the first time in MI actually appear to be fairly common. *Pycnopsyche circularis*, in particular, was found at the majority of Upper Peninsula sites collected during September 2019. *Pycnopsyche aglona* was found in both the Upper and Lower Peninsulas. Lack of previous documentation of these species was almost certainly due to a lack of collecting during autumn. Similarly, *A. ladogensis* and *P. indecisa* were abundant in June, but absent in July, demonstrating the importance of sampling during the early flight season.

The paper also demonstrates the importance of continued collecting, even in a state as well-documented as MI. Twelve of the 15 species mentioned in this paper were found in the remote and poorly-collected Huron Mountains, among other places. Another 2 species were found exclusively in a neighborhood stream outside of metro Detroit. Since it is difficult to predict which habitats will contain new or unique species, the best approach is to collect from as many habitats as possible.

The state of MI is now known to contain 305 caddisfly species, notably more than the adjacent states of MN (277) and OH (270). While not perfectly equivalent, all 3 states have had an extensive recent collecting effort (Armitage et al. 2011, Houghton 2012). Considering that 67 new state record caddisflies have been documented in MI during the last 10 years (Houghton et al. 2011, DeWalt and South 2015, Houghton 2016, Houghton et al. 2018) additional unknown species probably remain to be discovered.

### Acknowledgments

Funding for this study was supported by a grant from the Huron Mountain Wildlife Foundation, a Faculty Summer Leave grant from Hillsdale College, and funds from the Hillsdale College biology department. I thank Erin Flaherty, Ryan Lardner, and Mary Clare Smith for their efforts in collecting many of the specimens reported herein. This is paper #23 of the GH Gordon BioStation Research Series.

### Literature Cited

**Armitage B.J., Harris S.C., Schuster G.A., Usis J.D., MacLean D.B., Foote B.A., Bolton M.J., Garono R.J. 2011.** Atlas of Ohio aquatic insects, Volume 1: Trichoptera. Ohio Biological Survey Miscellaneous Contributions, 13.

**Bright E., Bidlack D. 1998.** *Psilotreta indecisa* and *Agarodes distinctus* (Trichoptera: Odontoceridae, Sericostomatidae): new state records and notes on the habitat of these species in Michigan. The Great Lakes Entomologist 31: 199–200.

**Cushman R.M. 2014.** Biogeography of the caddisfly genus *Arctopsyche* McLachlan, 1868 (Trichoptera: Hydropsychidae) in North America. The Pan-Pacific Entomologist 90: 174–181.

**DeWalt R.E., South E.J. 2015.** Ephemeroptera, Plecoptera, and Trichoptera in Isle Royale National Park, USA, compared to mainland species pool and size distribution. Zookeys 532: 137–158.

**Ellis R.J. 1962.** Adult caddisflies (Trichoptera) from Houghton Creek, Ogemaw County, Michigan. Occasional papers of the Museum of Zoology, University of Michigan 624: 1–16.

**Givens D.R., Ruiter D.E. 2015.** Clarification of the taxonomic status and distribution of *Arctopsyche inermis* Banks, 1943 and *Arctopsyche ladogensis* (Kolenati, 1859) (Trichoptera: Hydropsychidae: Arctopsychinae). The Pan-Pacific Entomologist 91: 192–195.

**Houghton D.C. 2012.** Biological diversity of Minnesota caddisflies. ZooKeys Special Issues 189: 1–389.

**Houghton D.C. 2016.** The caddisflies (Trichoptera) of an undisturbed Lower Michigan habitat. The Great Lakes Entomologist 49: 41–54.

**Houghton D.C. 2018.** When to sample adult caddisflies: data from a five-year study of a

first-order Michigan (USA) stream. Journal of Freshwater Ecology 33: 211–221.

**Houghton D.C, Brandin C.M., Brakel K.A. 2011.** Analysis of the caddisflies (Trichoptera) of the Manistee River watershed, Michigan. Great Lakes Entomologist 44: 1–15.

**Houghton D.C., DeWalt W.E., Pytel A.J., Brandin C.M., Rogers S.R., Hudson P.L., Ruiter D.E., Bright E., Armitage B.J. 2018.** Updated checklist of the Michigan caddisflies (Insecta: Trichoptera) with regional and habitat affinities. ZooKeys 730: 55–72.

**Karl T.S., Hilenhoff W.L. 1979.** The caddisflies (Trichoptera) of Parfrey's Glen Creek, Wisconsin. Wisconsin Academy of Science, Arts, and Letters 67: 31–42.

**Leonard J.W., Leonard F.A. 1949.** An annotated list of Michigan Trichoptera. Occasional papers of the Museum of Zoology, University of Michigan 522: 1–36.

**MacLean D.B. 1995.** Adult Trichoptera of the Devil Track River Watershed, Cook County, Minnesota and their role in biomonitoring. Great Lakes Entomologist 28: 135–154.

**Rasmussen A.K., Morse J.C. 2018.** Distributional checklist of Nearctic Trichoptera (August 2018 Revision). Unpublished, Florida A&M University, Tallahassee. 506 pp. [Available at http://www.trichoptera.org].

**Schmude K.L, Hilsenhoff W.L. 1986.** Biology, ecology, larval taxonomy, and distribution of Hydropsychidae (Trichoptera) in Wisconsin. The Great Lakes Entomologist 19: 123–145.

**Woods K.D. 2015.** An all-taxa biodiversity inventory of the Huron Mountain Club. Version January 2015. Occasional papers of the Huron Mountain Wildlife Foundation, No. 5.

**Yanoviak S.P., McCafferty W.P. 1996.** Comparison of macroinvertebrate assemblages inhabiting pristine streams in the Huron Mountains of Michigan, USA. Hydrobiologia 330: 195–211.

# Hunting Billbug *Sphenophorus venatus* (Coleoptera: Curculionidae: Dryophthorinae) Adult Feeding Preference and Attraction to Warm- and Cool-Season Turfgrasses

Alexandra G. Duffy[1,2,*] and Douglas S. Richmond[1]

[1] Purdue University, Department of Entomology, 901 West State Street, West Lafayette, IN 47907
[2] Brigham Young University, Department of Biology, 4102 Life Science Building, Provo, UT 84602
[*] Corresponding author: (e-mail: agduffy4@gmail.com)

**Abstract**

The hunting billbug *Sphenophorus venatus* Say (Coleoptera: Curculionidae: Dryophothorinae) is a generalist stem-boring pest on warm- and cool-season grasses. The objectives of this work were to (1) investigate adult feeding preference for four common turfgrass host species and (2) explore whether adults are attracted to the volatile odors emitted by these grasses. In laboratory feeding assays, *S. venatus* adults preferred zoysiagrass *Zoysia japonica* Steud 'Meyer' over all other species tested: Bermudagrass *Cynodon dactylon* 'Patriot', Kentucky bluegrass *Poa pratensis* 'Barron', and creeping bentgrass *Agrostis stolonifera* 'Penncross'. In y-tube olfactometry assays, only males were attracted to bermudagrass. Our findings suggest that although *S. venatus* odor recognition potentially influence dispersal and host-seeking behavior, it may not ultimately determine feeding preference, where more close-range cues may be important. Further investigations on the mechanisms that cause variation in these behaviors could aid in the pursuit of more sustainable management techniques, such as strategic plant species selection and development of synthetic semiochemical lures for monitoring and trapping.

**Keywords:** olfactometry, integrated pest management

---

Beetles in the genus *Sphenophorus* Schönherr, commonly referred to as billbugs, are stem-boring, cosmopolitan pests of small grain crops and grasses (Vaurie 1951). A particularly problematic billbug species in urban environments is the hunting billbug, *Sphenophorus venatus* Say (Coleoptera: Curculionidae: Dryophthorinae), which causes severe injury to warm-season (C-4) grasses, such as zoysiagrass (*Zoysia* spp. Willd.) and bermudagrass (*Cynodon* spp. Pers.) (Doskocil and Brandenburg 2012, Huang and Buss 2013, Reynolds et al. 2016), but also damages cool-season (C-3) grasses, such as tall fescue (*Festuca arundinacea* Schreb.), Kentucky bluegrass (*Poa pratensis* L.), and creeping bentgrass (*Agrostis stolonifera* L. var. *palustris* Huds.) (Johnson-Cicalese and Funk 1990, Johnson-Cicalese et al. 1990, Vittum et al. 1999). Because of their host breadth, the hunting billbug is a particularly problematic pest in lawns and other urban greenspaces where both warm- and cool-season turfgrasses are grown (Milesi et al. 2005), and synthetic insecticides are often used to control their damage (Williamson et al. 2013). Because of public concern surrounding the use of chemical pesticides in these environments (Johnson et al. 2013), billbugs are one of a handful of insects that persist as a major obstacle for municipalities looking to reduce chemical inputs and create more sustainable recreational spaces. Adult billbugs chew notches in grass tillers and then females oviposit directly into the tiller (Williamson et al. 2013), thus infestations of the damaging larval stage are driven largely by adult host preferences. Understanding adult feeding behavior could help promote sustainability in urban turfgrass systems by identifying grass species that are less attractive to this pest and that, in turn, require fewer pesticides. We conducted two laboratory studies to 1) investigate the feeding preference of hunting billbug adults for warm- vs. cool-season grasses and 2) explore whether hunting billbug adults are attracted to the odors of different grass species.

## Materials and Methods

**Insect and Plant Material.** *S. venatus* adults were collected from bermudagrass *C. dactylon* 'Patriot' athletic fields in West Lafayette, IN from May to October 2015 using pitfall traps or hand collecting at night. Billbugs were separated by sex (Johnson-Cicalese et al. 1990) and maintained in a growth chamber (25–27 °C; 78–85% RH; 10:14 (L:D)) in plastic deli cups (12 cm diameter) with a moist paper towel and clippings of bermudagrass host-plant

material until used in bioassays (12 to 48 hours). Individuals were used only once, and the same individuals were not used in both feeding choice and olfactometry assays. Two cool-season grasses, Kentucky bluegrass (*P. pratensis* 'Park') and creeping bentgrass (*A. stolonifera* 'Penncross'), and two warm-season grasses, bermudagrass (*C. dactylon* 'Patriot') and zoysiagrass (*Z. japonica* 'Meyer'), from plots at the Purdue University W.H. Daniel Center for Turfgrass Research and Diagnostics were used for plant material in both assays.

**Feeding choice assays.** Adult feeding preference was tested using a series of petri-dish bioassays adapted from Richmond (1999) and Johnson-Cicalese and Funk (1990). The bioassays incorporated two cool-season grasses, Kentucky bluegrass (*P. pratensis* 'Park') and creeping bentgrass (*A. stolonifera* 'Penncross'), and two warm-season grasses, bermudagrass (*C. dactylon* 'Patriot') and zoysiagrass (*Z. japonica* 'Meyer'). Binary choice treatments included: 1) bermudagrass vs. Kentucky bluegrass, 2) bermudagrass vs. bentgrass, 3) zoysiagrass vs. Kentucky bluegrass, 4) zoysiagrass vs. bentgrass, and 5) bermudagrass vs. zoysiagrass. Treatments encompassed all combinations of warm- vs. cool-season grasses as well as a comparison of the two warm-season grasses. Each treatment was replicated 20 times for males and 20 times for females.

Adult billbugs were placed individually in 5 cm petri dishes lined with moist filter paper and isolated with no food for 24 hours before providing fresh grass tillers. One tiller of each grass species was provided to each billbug and petri dishes were arranged on the bench in a randomized complete block design. Feeding scars were used as an indicator of preference (Richmond 1999) and were counted using an Olympus IX51 dissecting microscope after 24, 48, and 72 hours. New grass tillers were provided at the beginning of each 24-hour interval. The influence of grass species and insect sex on variation in the mean number of feeding scars were tested for each of the binary-choice treatments using analysis of variance (ANOVA) followed by Tukey's HSD (Statistica 13, Dell Inc. 2016).

**Olfactometry.** The behavioral response of *S. venatus* adults to the odors from four grass species (Kentucky bluegrass, creeping bentgrass, bermudagrass, and zoysiagrass) was tested using a y-tube olfactometer (8 cm and 12 cm arms; 2 cm diameter, round glass joints; Analytical Research Systems, Gainesville, FL), a well-developed and widely used method for quantifying insect responses to plant odors. Odor stimuli were delivered by pushing purified air through an activated charcoal filter and split into two air steams (1 L/min) which were blown through a glass container (14 cm long, 2 cm diameter) that was either empty (purified air control) or contained 5 grams of above-ground plant clippings from one of the four grass species. The odors were then delivered to one of the arms of the olfactometer. Glassware was washed using 1% diluted Alconox soap, rinsed with acetone, and baked in an oven for ~10 minutes at 200 °C between observations. Each odor treatment was compared to a purified air control and fresh odor sources were used for no longer than 30 minutes (McGraw et al. 2011). All observations were made between 9:00 PM and 6:00 AM using a red headlamp. Based on Duffy et al. (2018), a billbug was considered responsive if it walked upwind in the 12-cm base arm and then turned to walk 2 cm into one of the 8–cm arm corresponding with an odor source (treatment or control) and remained within that arm for at least one minute. Billbugs not making a choice between the two odor treatment arms within ten minutes were considered non-responsive (McGraw et al. 2011, Duffy et al. 2018). Each individual billbug was used once, and treatment arms were switched after each replicate to control for directional bias. Treatments were replicated until 20 responsive male and female adults were observed. A Chi-square goodness of fit test was used to test the null hypothesis that *S. venatus* adults showed no preference for the host-plant odor source vs. purified air control (Statistica 13, Dell Inc. 2016).

## Results

**Feeding choice assays.** For warm- vs. cool-season grass treatments, the interaction between grass species and billbug sex was not significant ($F \leq 1.57$; df = 1, 76; $P \geq 0.215$). Grass species did not have a significant effect on the mean number of adult feeding scars for bermudagrass vs. Kentucky bluegrass ($F = 0.03$; df = 1, 72; $P = 0.869$; Fig. 1A) or bermudagrass vs. bentgrass ($F = 0.30$; df = 1, 76; $P = 0.585$; Fig. 1B). Grass species did, however, affect the mean number of feeding scars observed for both the zoysiagrass vs. Kentucky bluegrass ($F = 10.81$; df = 1, 76; $P = 0.002$; Fig. 1C), and zoysiagrass vs. bentgrass treatments ($F = 34.70$; df = 1, 74; $P < 0.001$; Fig. 1D), with zoysiagrass having a higher amount of feeding in both cases. In addition, sex affected the number of feeding scars for the zoysiagrass vs. bentgrass treatment ($F = 15.33$; df = 1, 74; $P < 0.001$; Fig. 1D), with males feeding more on both grass species than females.

When given a choice between the two warm-season grasses, bermudagrass and zoysiagrass, the number of feeding scars

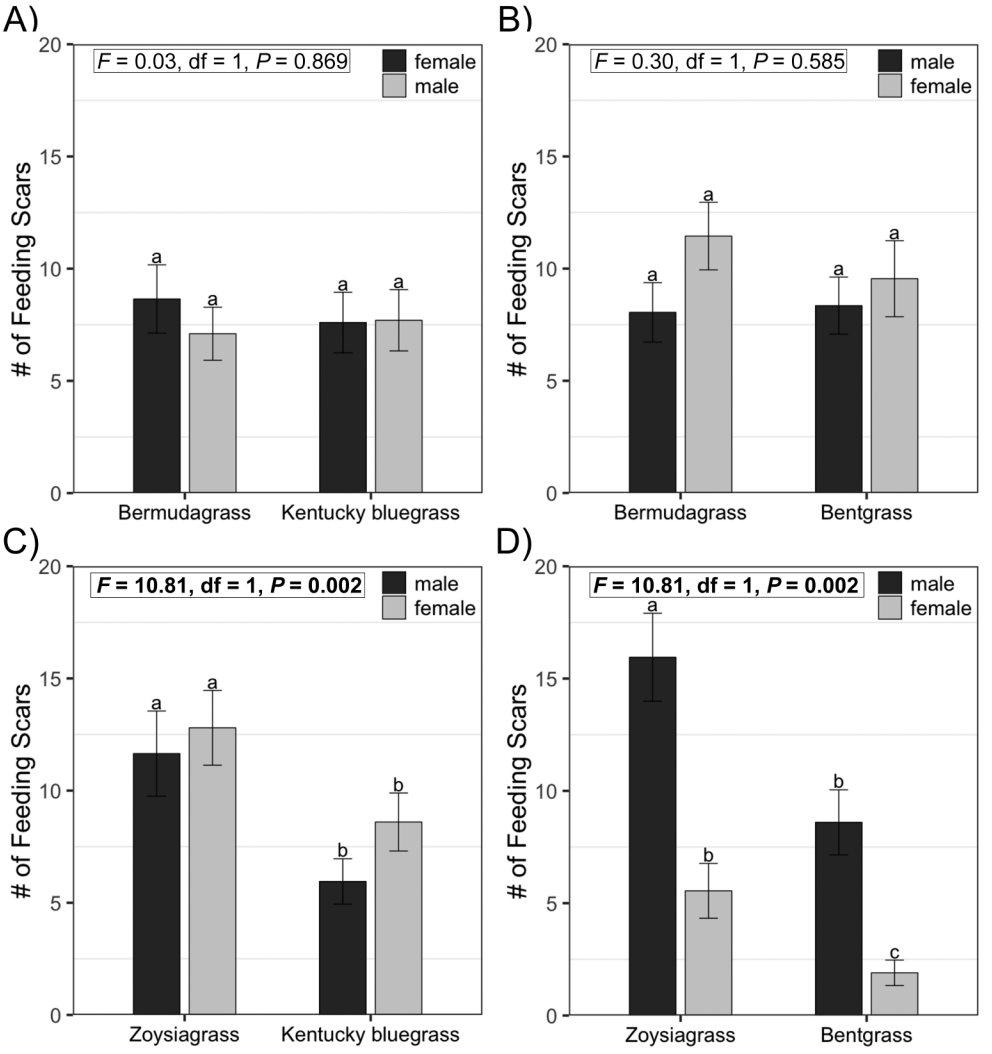


**Figure 1.** Number of hunting billbug *Sphenophorus venatus* Say feeding scars on warm-season turfgrass, bermudagrass (A and B) and zoysiagrass (C and D) vs. cool-season turfgrass, Kentucky bluegrass (A and C) and bentgrass (B and D) in laboratory choice bioassays (n = 40). Differences were tested with analysis of variance (ANOVA) followed by Tukey's HSD. Because there was no significant interaction between 'grass species' and 'sex', the F and p-values represent the effect of 'grass species' and significant effects ($P < 0.05$) are indicated in bold. Means with the same letter are not significantly different ($P > 0.05$).

varied with grass species and billbug sex (grass species × sex interaction, $F = 16.08$; df = 1, 76; $P < 0.001$), with males feeding more on zoysiagrass than bermudagrass whereas females exhibited no preference between the two warm-season grass species (Fig. 2).

**Olfactometry.** Across all treatments, 79.7% (n = 192) of the billbugs tested were responsive and walked upwind in the y-tube olfactometer and made a choice between odor treatments. Adult *S. venatus* males were attracted to bermudagrass in y-tube olfactometry bioassays ($X^2 = 5.00$; df = 1; $P = 0.025$), with 75% (n = 20) of males choosing the arm with the odor treatment containing the bermudagrass host-plant material relative to a purified air control. In contrast, females were not attracted to bermudagrass ($X^2 = 0.80$; df = 1; $P = 0.317$). Neither males nor females were attracted to the odors from any of the other host species, zoysiagrass ($X^2 \leq 1.80$; df = 1; $P \geq 0.180$), Kentucky bluegrass

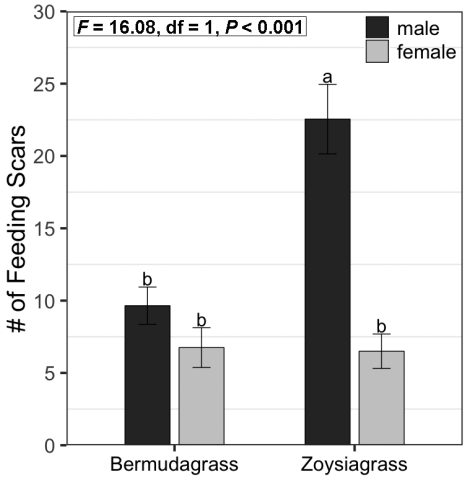


**Figure 2.** Number of hunting billbug, *Sphenophorus venatus* Say, feeding scars on bermudagrass vs. zoysiagrass from males (n = 20) and females (n = 20) in laboratory choice bioassays. Differences were tested with analysis of variance (ANOVA) followed by Tukey's HSD. *F* and *P*-value represent the interaction effect of 'grass species' × 'sex'. Means with the same letter are not significantly different ($P > 0.05$).

($X^2 \leq 0.80$; df = 1; $P \geq 0.371$), or bentgrass ($X^2 \leq 0.80$; df = 1; $P \geq 0.371$) compared to the purified air control.

## Discussion

Information on hunting billbug feeding behavior is limited (Johnson-Cicalese and Funk 1990, Reinert et al. 2011, Huang and Buss 2013). This is the first study to investigate feeding preference of billbugs when provided a choice between different grass species, and the first to survey multiple species of both warm- and cool-season grass types. *Sphenophorus venatus*, displayed a preference for one specific warm-season species, zoysiagrass, feeding more on this species when compared to any of the other grass species examined. This initial study suggests that the other grasses may be less preferred habitats for this species, which may reduce the likelihood of damage and subsequently reduce reliance on synthetic insecticides. However, additional field experiments, such as measuring the feeding and damage rates within monoculture turf stands, should be conducted before drawing definitive conclusions on selecting these grasses as a management strategy. Future work should also include additional warm- and cool-season grass species to the ones tested in this study, as well as multiple genotypes or cultivars within those species (Reinert et al. 2011, Huang and Buss 2013).

Host-seeking behavior directed by plant odors serves as a more energy efficient strategy for locating preferred hosts than random foraging. The ability for *S. venatus* to orient towards odors was first documented by Duffy et al. (2018), but this study only tested a single grass species, bermudagrass. The present study assessed the attractiveness of four different grass species, Kentucky bluegrass, creeping bentgrass, bermudagrass, and zoysiagrass (*Z. japonica* 'Meyer'). We observed that male, but not female, hunting billbug adults were attracted to bermudagrass in the y-tube olfactometer. Surprisingly, billbugs were not attracted to any of the other grass species tested, even though zoysiagrass was their preferred host in feeding assays. These results are intriguing, as volatile attraction is known to be influenced by previous host exposure in other beetle species (Austel et al. 2014). The billbugs in this study were collected from a well-established population infesting a bermudagrass monoculture and were only attracted to bermudagrass in the olfactometer. It would be interesting to test for the olfactory responses of individuals from other populations and geographical regions which colonize different grass species and investigate if a pattern of requiring previous exposure to elicit attractiveness would emerge. Future research on intraspecific variation in the attraction to odors and the potential influence of feeding experience could provide insight into the utility of regional or even site specific semiochemical lures for monitoring and management, as well as the ability for these pests to colonize novel host plants. Lack of attraction towards the odors of any grasses by female hunting billbugs could be due to reliance on a putative male-produced pheromone hypothesized by Duffy et al. (2018). It would be interesting to investigate whether the addition of conspecifics alters the attractiveness of grass species in a future study. The observations from this study suggest that a species-specific blend of odors may be unique to bermudagrass and that this odor blend is particularly attractive to this population of *S. venatus* males. In pursuit of more sustainable monitoring and management techniques, such as synthetic semiochemical lures, future efforts should focus on isolating and identifying the specific volatile compounds or blend of compounds that elicited the observed attractiveness.

Our findings from both the feeding preference and olfactometry assays suggest plant selection may be a useful strategy in urban landscape managers' efforts to create more sustainable urban environments that are less vulnerable to insect pests. Although

it appears male hunting billbugs are attracted to the odor of bermudagrass, they displayed a clear feeding preference for zoysiagrass. However, the cool-season grasses (Kentucky bluegrass and creeping bentgrass) were less preferred in both feeding and olfactometry assays by both males and females. Our findings imply that these grasses may be good candidates for creating urban habitats that are less attractive and less preferred by hunting billbugs. Field studies are still needed to test this hypothesis, but our findings suggest it may be possible to reduce the amount of feeding and resultant damage in urban greenspaces vulnerable to hunting billbug infestation by planting cool-season grasses, such as Kentucky bluegrass and creeping bentgrass.

## Acknowledgments

We thank Danielle Craig and Garrett Price for field assistance. We are grateful for the cooperation of superintendents and staff at Purdue Sports Turf and William H. Daniel Turfgrass Research and Diagnostic Center. We are grateful for the cooperation of Dr. Ian Kaplan and Ulianova Vidal Gómez for assistance with the olfactometry methods. Dr. Juli Carrillo, Dr. Laura Ingwell, and Dr. Elizabeth Long provided helpful insights on earlier versions of this manuscript. Dr. Diane Silcox Reynolds provided helpful insight and was integral in developing the initial questions during the A. Duffy's undergraduate research project. Funding was provided by the United States Golf Association (2015-10-525).

## Literature Cited

**Austel, N., C. Bjorkman, M. Hiker, and T. Meiners. 2014.** Phenotypic plasticity in host plant preference of the willow leaf beetle *Phratora vulgatissima*: the impact of experience made by adults. Agricultural and Forest Entomology 16: 417–425.

**Dell Inc. 2016.** Dell Statistica (data analysis software system), version 13. software.dell.com.

**Doskocil, J. P., and R. L. Brandenburg. 2012.** Hunting billbug (Coleoptera: Curculionidae) life cycle and damaging life stage in North Carolina, with notes on other billbug species abundance. Journal of Economic Entomology 1105: 2045–2051.

**Duffy, A. G., G. P. Hughes, M. D. Ginzel, and D. S. Richmond. 2018.** Volatile and contact chemical cues associated with *Sphenophorus venatus* and *Sphenophorus parvulus* (Coleoptera: Dryophthoridae) host and mate recognition behavior. Journal of Chemical Ecology 44: 556–564.

**Huang, T. I., and E. A. Buss. 2013.** *Sphenophorus venatus vestitus* (Coleoptera: Curculionidae) preference for bermudagrass cultivars and endophytic perennial ryegrass overseed. Florida Entomologist 96(4): 1628–1630.

**Johnson, P. G., F. S. Rossi and B. Horgan. 2013.** Sustainable turfgrass management in an increasingly urbanized world. pp. 1007–1028 In: Turfgrass: Biology, Use and Management (J. C. Stier, B.P. Horgan and S. A. Bonos eds.), Agronomy Monograph 56, American Society of Agronomy, Crop Science Society of America, Soil science Society of America, Madison, WI, USA.

**Johnson-Cicalese, J. M., and C. R. Funk. 1990.** Additional host plants of four species of billbug found on New Jersey turfgrasses. Journal of the American Society for Horticultural Science 115: 608–611.

**Johnson-Cicalese, J. M., G. W. Wolfe, and C. R. Funk. 1990.** Biology, distribution, and taxonomy of billbug turf pest (Coleoptera: Curculionidae). Environmental Entomology 19: 1037–1046.

**McGraw, B. A., C. Rodriguez-Saona, R. Holdcraft, Z. Szendrei, and A. M. Koppenhöfer. 2011.** Behavioral and electrophysiological responses of *Listronotus maculicollis* (Coleoptera: Curculionidae) to volatiles from intact and mechanically damaged annual bluegrass. Environmental Entomology 40: 412–19.

**Milesi, C., S. W. Running, C. D. Elvidge, J. B. Dietz, B. T. Tuttle, and R. R. Nemani. 2005.** Mapping and modeling the biogeochemical cycling of turf grasses in the United States. Environmental Management 36: 426–438.

**Reinert, J., M. Engelke, and J. Heitholt. 2011.** Hunting billbug (Coleoptera: Curculionidae) resistance among zoysiagrass (*Zoysia spp.*) cultivars. Florida Entomologist 94: 613–621.

**Reynolds, D. S., W. C. Reynolds, and R. L. Brandenburg. 2016.** Overwintering, oviposition, and larval survival of hunting billbugs (Coleoptera: Curculionidae) and implication for adult damage in North Carolina turfgrass. Journal of Economic Entomology 109: 240–248.

**Richmond, D. S. 1999.** Influence of endophytic perennial ryegrass (*Lolium perenne*) on the population density of two insects and plant species composition in mixed turfgrass swards. Ph.D. Dissertation, The Ohio State University.

**Vaurie, P. 1951.** Revision of the genus *Calendra* (formerly *Sphenophorus*) in the United States and Mexico (Coleoptera: Curculionidae). Bulletin of the American Museum of Natural History 93: 33–186.

**Vittum, P., M. Villani, and H. Tashiro. 1999.** Turfgrass in the Modern Environment. *In* Turfgrass Insects of the United States and Canada. Cornell University Press, Ithaca, NY, USA.

**Williamson, R. C., D. W. Held, R. Brandenburg and F. Baxendale. 2013.** Turfgrass Insect Pests. Pp 809–890 In: Turfgrass: Biology, Use and Management (J. C. Stier, B.P. Horgan and S. A. Bonos eds.), Agronomy Monograph 56, American Society of Agronomy, Crop Science Society of America, Soil science Society of America, Madison, WI, USA.

# *Karshomyia caulicola* (Diptera: Cecidomyiidae) Associated with *Sclerotinia*-Infected Soybean in the United States and Canada

Robert L. Koch[1,*], Bruce D. Potter[2], Joseph Moisan-De Serres[3], Janet J. Knodel[4], Veronica Calles-Torrez[4], John Gavloski[5], Theresa Cira[1], Mads Bartz[1], Raymond J. Gagné[6]

[1] Department of Entomology, University of Minnesota, 1980 Folwell Avenue, Saint Paul, MN 55108
[2] Southwest Research and Outreach Center, University of Minnesota Extension, Lamberton, MN 56152
[3] Laboratoire d'Expertise et de Diagnostic en Phytoprotection, Ministère de l'Agriculture, des Pêcheries et de l'Alimentation du Québec, QC, Canada
[4] Extension Entomology, Department of Plant Pathology, North Dakota State University, Walster Hall, Fargo, ND 58102
[5] Manitoba Agriculture, Box 1149, 65-3rd Avenue NE, Carman, MB, Canada
[6] Systematic Entomology Laboratory, Agricultural Research Service, U.S. Department of Agriculture, c/o Smithsonian Institution MRC-168, P.O. Box 37012, Washington, DC 20013-7012, USA
[*] Corresponding author: (e-mail: koch0125@umn.edu)

**Abstract**

The white-mold gall midge, *Karshomyia caulicola* (Coquillett), (Diptera: Cecidomyiidae) was documented in association with soybean, *Glycine max* (L.) Merr., infected with the fungus *Sclerotinia sclerotiorum* (Lib.) de Bary. This mycetophagous cecidomyiid appears widespread in the northern soybean producing region, with confirmed detections from Minnesota, North Dakota (U.S.) and Québec (Canada). Though likely not a pest of soybean plants, the presence of *K. caulicola* in soybean fields may complicate identification, population assessment and decision making for soybean gall midge, *Resseliella maxima* Gagné, which is a recently described pest of soybean. Here, we provide an overview of the known biology and distribution of *K. caulicola* and descriptions to aid in distinguishing these two cecidomyiids.

**Keywords:** White mold, *Sclerotinia* stem rot, Cecidomyiidae, gall midge

---

This article documents the association of the white-mold gall midge, *Karshomyia caulicola* (Coquillett) (Diptera: Cecidomyiidae), with soybean, *Glycine max* (L.) Merr. (Fabales: Fabaceae), and other plants infected with the fungus *Sclerotinia sclerotiorum* (Lib.) de Bary (Helotiales: Sclerotiniaceae). This fungus causes the plant disease *Sclerotinia* stem rot (white mold), with infected plants exhibiting growth of white, cottony mycelia and sclerotia on and in above-ground tissues (Link and Johnson 2007). Of particular concern is that immature stages of *K. caulicola* on *Sclerotinia*-infected soybean plants or in soil of soybean fields could be confused for immatures of the recently described soybean gall midge, *Resseliella maxima* Gagné (Diptera: Cecidomyiidae) (Gagné et al. 2019). *Resseliella maxima* is an emerging pest of soybean in the Midwest Region of the U.S. (Gagné et al. 2019). *Resseliella maxima* infests the stems of soybean plants generally near the soil surface, where the larvae feed mainly on the phloem, but may eventually move to the xylem and pith (Gagné et al. 2019, Koch and Potter 2019).

During surveys to determine the geographic distribution of *R. maxima* and other scouting in soybean, larvae with a superficially similar appearance to *R. maxima* were found on or in *Sclerotinia*-infected tissues of soybean plants. Cecidomyiid adults were reared under ambient laboratory conditions from larvae in *Sclerotinia*-infected soybean stems collected in Val-Saint-François Municipality, Québec (Canada) (29 August 2018) and Ramsey County, Minnesota (U.S.) (2 August 2019). In addition, two cecidomyiid adults were collected in an emergence trap placed over the soil in a field in Stearns County, Minnesota (U.S.) (8 July 2019). In 2018, this field had a significant infestation of cecidomyiid larvae associated with *Sclerotinia*-infected soybean stems. Additional cecidomyiid larvae were collected from *Sclerotinia*-infected soybean stems Redwood (17 September 2019), Renville (16 September 2019) and Stearns (11 September 2019) counties in Minnesota, and in Barnes (17 September 2019), Cass (17 September 2019), LaMoure (17 September 2019), Richland (17, 20 and 29 September 2019) and Sargent (17 September 2019) counties in North Dakota

(U.S.). The above-mentioned cecidomyiid adults and larvae from Québec, Minnesota and North Dakota were determined to be *K. caulicola* by R. J. Gagné. Voucher specimens are deposited in the National Museum of Natural History (USNM), Smithsonian Institution, Washington, DC (U.S.).

Larvae matching the description of *K. caulicola* were reported (but not brought to the laboratory for identification) from *Sclerotinia*-infected tissues of soybean plants in 2018 in McLeod, Sibley, Stearns, Olmsted, Ottertail and Wadena counties in Minnesota (Potter unpublished data). In 2019, more larvae were reported from *Sclerotinia*-infected tissues of soybean plants in Benton, Brown, Chippewa, Dakota, Douglas, Goodhue, Kandiyohi, Meeker, Morrison, Nicollet, Ottertail, Pope, Redwood, Ramsey, Sherburne, Stearns, Stevens, Wadena, Waseca and Wright counties in Minnesota (Potter and Koch unpublished data), and from *Sclerotinia*-infected pepper, *Capsicum annuum* L. (Solenales: Solenaceae), in Val-Saint-François Municipality, Québec (Moisan-De Serres personal observation). Previously, observations were made of similar larvae in association with *Sclerotinia*-infected tissues of soybean, dry beans, *Phaseolus vulgaris* L. (Fabales: Fabaceae), and canola, *Brassica napus* L. (Brassicales: Brassicaceae), in Manitoba (Canada) in 2014 and 2016 (Gavloski and Bajracharya 2016, Gavloski unpublished data), and in association with *Sclerotinia*-infected soybean and sunflower, *Helianthus annuus* L. (Asterales: Asteraceae), in Minnesota dating back at least 25 years (Potter personal observation).

These larvae associated with *Sclerotinia*-infected tissues displayed a distinctly different biology than those of typical *R. maxima* infestations (Gagné et al. 2019, Koch and Potter 2019). Infestations of *K. caulicola* were seen later in the season (after soybean flowering and the onset of *Sclerotinia* stem rot), whereas *R. maxima* infestations are typically found as early as the third trifoliate stage growth stage of soybean. *Karshomyia caulicola* larvae were found inside and outside of *Sclerotinia*-infected soybean stems, pods and other tissues (Fig. 1), whereas *R. maxima* larvae are typically found under the epidermis of the stem at the base of soybean plants. *Karshomyia caulicola* infestations were encountered throughout soybean fields wherever *Sclerotinia*-infected stems are present, whereas *R. maxima* infestations have typically been most abundant near field edges that are adjacent to fields that had soybean the previous year.

In the field, late instar larvae of *K. caulicola* displayed a generally less intense orange coloration (Fig. 1) than similarly sized larvae of *R. maxima* (Gagné et al. 2019). Under close examination, larvae of these two species can be distinguished. The larval integument of *K. caulicola* is generally smooth with minor sculpturing (Fig. 2a). The terminal abdominal segment of *K. caulicola* ends in two blunt lobes, one on each side, and each with four inconspicuous apical setae (Fig. 2a, b). The spatula of *K. caulicola* is deeply incised anteriorly and

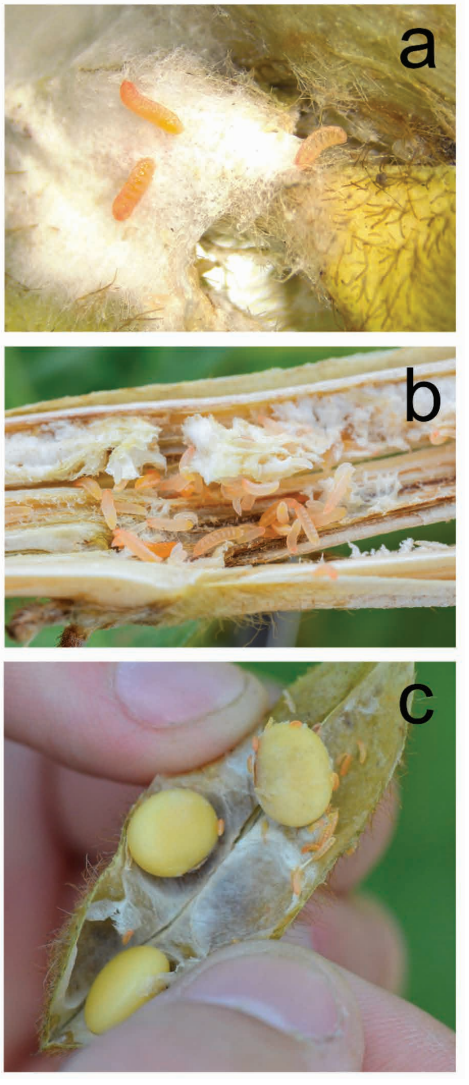


**Figure 1:** Larvae of *Karshomyia caulicola* associated with *Sclerotinia*-infected soybean: larvae on mycelia on exterior of soybean pods [a], larvae inside a *Sclerotinia*-infected stem [b], and larvae inside a *Sclerotinia*-infected pod [c]. Image by T. Cira & R. Aita [a], J. Moisan-De Serres [b] and B.D. Potter [c].

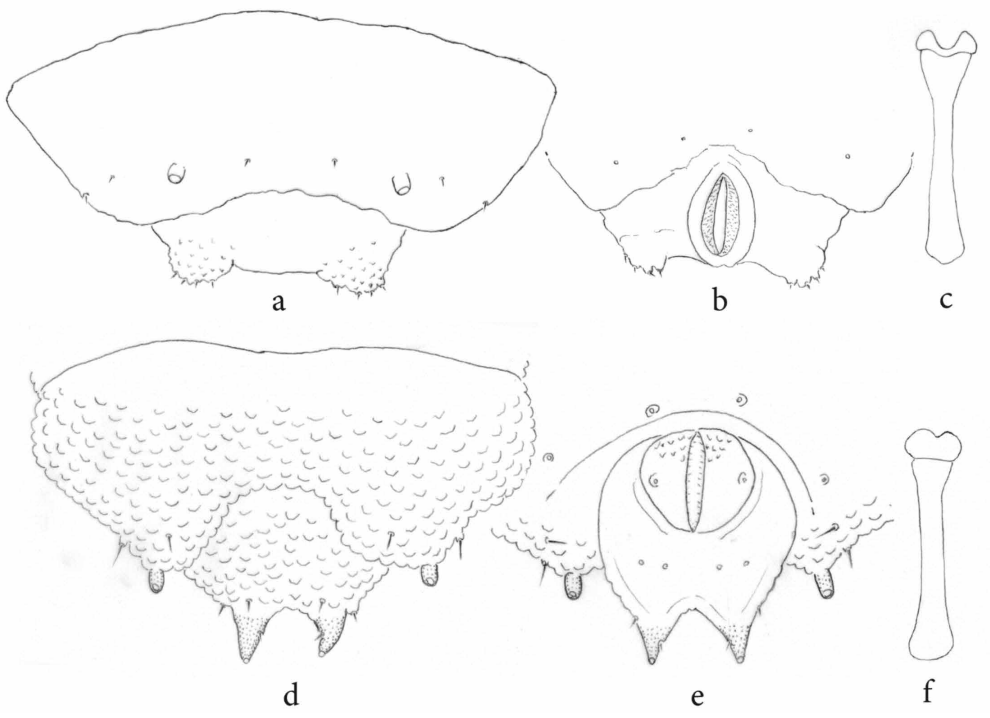


**Figure 2:** Distinguishing characters of *Karshomyia caulicola* larva (dorsal [a] and ventral [b] views of larval eighth and terminal abdominal segments and spatula [c]) and *Resseliella maxima* larva (dorsal [d] and ventral [e] views of larval eighth and terminal abdominal segments and spatula [f]). Drawings by R.J. Gagné.

the lobes rounded (Fig. 2c). In contrast, the larva of *R. maxima* has a uniformly pebbled integument, and its terminal segment is more tapered and ends in a conspicuous pair of conical projections (Fig. 2d, e). The spatula of *R. maxima* is only weakly incised apically (Fig. 2f).

Adults of the two species are strikingly different. Adults of *K. caulicola* have uniformly grayish antennae, wings and legs, and the abdominal tergites and sternites are very definitely subdivided horizontally (Fig. 3). In contrast, adults of *R. maxima* have striped antennae and legs, mottled wings, and the abdominal tergites and sternites are uniformly sclerotized (Gagné et al. 2019). The male sexual apparatus of both are distinct also; that of *K. caulicola* was illustrated in Gagné (1973) and that of *R. maxima* in Gagné et al. (2019). A key to genera of adult Cecidomyiinae of North America is available (Gagné 2018) to identify other possible gall midges that might be encountered in work on soybean.

The genus *Karshomyia*, which is comprised of 51 species worldwide, is in the tribe Karshomyiini in the subfamily Cecidomyiinae (Gagné and Jaschhof 2017). *Karshomyia caulicola* has also been referred to as: *Diplosis caulicola* Coquillett, *Hiastatus concinnus* Marikovskij, and *Karschomyia concinna* (Marikovskij) (Pakalniškis et al. 2000, Gagné and Jaschhof 2017). *Karshomyia caulicola* has a Holarctic distribution. It is widespread in Europe, with reports from the United Kingdom (Harris 2004), Netherlands (Nijveldt 1985), Germany (Meyer 1984), Russia (Marikovskij 1956), Latvia (Spungis 2003), Lithuania (Pakalniškis et al. 2000) and Ukraine (Gagné and Jaschhof 2017). In Germany, *K. caulicola* is reported as univoltine with adult flight activity occurring over a period of five months (Meyer 1984). In North America, *K. caulicola* was previously reported from only the U.S. state of New Hampshire (Coquillett 1895). The detections reported above, expand considerably the known North American range of *K. caulicola*.

*Karshomyia caulicola* is reported as being mycetophagous (Meyer 1984), which explains the observed association with *Sclerotinia*-stem rot of plants. In Europe, larvae of this species have been reported from *Sclerotinia*-infected "stems of beans and potatoes and in the stems and pods of

**Figure 3:** Adult of *Karshomyia caulicola*. Image by J. Moisan-De Serres.

winter rape" and to feed on the mycelia of such fungi (Nijveldt 1985). Similarly, the detections reported here of *K. caulicola* in North America were also associated with *Sclerotinia*-infected stems and reproductive tissues of plants. Therefore, we suspect this species is not a plant pest. However, the report of *K. caulicola* larvae from stems of Icelandic poppy, *Papaver nudicaule* L. (Ranunculales: Papaveraceae), in New Hampshire (Coquillett 1895) and "oil poppy" in the United Kingdom (Harris 2004) had no mention of *Sclerotinia*-stem rot or other fungal infection of the plants.

Here, we provided the first documentation of *K. caulicola* associated with *Sclerotinia*-infected soybean and other plants in North America, which corroborates reports of its biology from Europe. The geographic extent of the reports presented here and a report from Pierce County, Wisconsin (U.S.) (Hamilton 2019) confirm that *K. caulicola* is widespread. Furthermore, knowledge of the presence of this cecidomyiid in soybean refines our understanding of the geographic distribution of *R. maxima*, which was previously confused with *K. caulicola* in some earlier distribution maps. The detection of adult *K. caulicola* in emergence cages placed in a field in 2019 that had numerous orange-colored cecidomyiid larvae in association with *Sclerotinia*-infected soybean in 2018, suggests *K. caulicola* likely overwinters in the soil or plant debris in the field. Additionally, we clarify the morphological differences between *K. caulicola* and *R. maxima* to facilitate identification of these species for efforts to understand the biology of both these species in soybean fields. The presence of both cecidomyiid species in soybean may complicate identification, population assessment and decision making for the plant pest, *R. maxima*.

## Acknowledgments

Some of the survey efforts that resulted in detections of *Karshomyia caulicola* were partially supported by the North Central Soybean Research Program, the Minnesota Soybean Research and Promotion Council, and the North Dakota Soybean Council.

## Literature Cited

**Coquillett, D. W. 1895.** Two dipterous insects injurious to flowers. Insect Life 7: 399–402.

**Gagné, R. J. 1973.** A review of *Karschomyia* Felt with descriptions of seven new Nearctic species (Diptera: Cecidomyiidae). Proceedings of the Entomological Society of Washington 75: 345–354.

**Gagné, R. J. 2018.** Key to adults of North American genera of the subfamily Cecidomyiinae (Diptera: Cecidomyiidae). Zootaxa 4392: 401–457.

**Gagné, R. J., and M. Jaschhof. 2017.** A Catalog of Cecidomyiidae of the World. 4th Edition. 762 pp. Available from https://www.ars.usda.gov/ARSUserFiles/80420580/Gagne_2017_World_Cat_4th_ed.pdf.

**Gagné, R. J., J. Yukawa, A. K. Elsayed, and A. J. McMechan. 2019.** A new pest species of *Resseliella* (Diptera: Cecidomyiidae) on soybean (Fabaceae) in North America with a description of the genus. Proceedings of the Entomological Society of Washington 121(2): 168–177. Available from https://doi.org/10.4289/0013-8797.121.2.168

**Gavloski, J., and P. Bajracharya. 2016.** Manitoba Insect & Disease Update, issue 14 (17 August 2016). Manitoba Agriculture. Available from http://www.manitoba.ca/agriculture/crops/seasonal-reports/insect-report-archive/print,insect-report-2016-08-17.html

**Hamilton, K. 2019.** White mold gall midge. Wisconsin Pest Bulletin. 7 November 2019. Available from https://datcpservices.wisconsin.gov/pb/pests.jsp?categoryid=5&articleid=3243&issueid=337

**Harris, K. M. 2004.** New records of Cecidomyiidae (Diptera) in Britain. Dipterist's Digest 11: 33–40.

**Koch, R. L., and B. D. Potter. 2019.** Soybean gall midge in Minnesota soybean. University of Minnesota Extension. Available from https://extension.umn.edu/soybean-pest-management/soybean-gall-midge-minnesota-soybean

**Link, V., and K. B. Johnson. 2007.** White mold. The plant health instructor. DOI: 10.1094/PHI-I-2007-0809-01.

**Marikovskij, P. I. 1956.** [New gall midges (Diptera, Itonididae) of the USSR.] Entomologicheskoe Obozrenie 35: 184–195.

**Meyer, H. 1984.** Experimentell-ökologische Untersuchungen an Gallmücken (Cecidomyiidae-Diptera) in Salzwiesenbereichen Nordwestdeutschlands. Faunistisch-Ökologische Mitteilungen, supplement 5: 1–124.

**Nijveldt, W. 1985.** Nieuwe galmuggen voor de Nederlandse fauna (IX) (Diptera: Cecidomyiidae). Entomologische Berichten 45: 85–88.

**Pakalniškis, S., J. Rimšaitė, R. Sprangauskaitė-Bernotienė, R. Butautaitė, and S. Podėnas. 2000.** Checklist of Lithuanian Diptera. Acta Zoologica Lituanica 10: 3–58.

**Spungis, V. 2003.** A checklist of Latvian Cecidomyiinae (Diptera, Cecidomyiidae) with notes on new records. Latvijas Entomologs 40: 5–11.

# First Record of the Hairy Maggot Blow Fly, *Chrysomya rufifacies* (Diptera: Calliphoridae) in Indiana

Kristi Bugajski*, PhD and Raenah Bailey

1610 Campus Drive East, Valparaiso University. Valparaiso, IN 46383.
* Corresponding author: (e-mail: Kristi.Bugajski@valpo.edu).

**Abstract**

The hairy maggot blow fly, *Chrysomya rufifacies* (Macquart 1844; Diptera: Calliphoridae) is native to Australia but has been spreading to a worldwide distribution and has become established in the United States. This paper presents the first documentation of the species in Indiana, specifically in Valparaiso, Indiana. The species was found in September 2017 and again in October 2019. Its presence is variable in Northwest Indiana, but it seems to be found in the fall when the temperatures are at least 30°C prior to collection.

**Key Words:** Hairy maggot, *Chrysomya rufifacies*, Indiana

---

The hairy maggot blow fly, *Chrysomya rufifacies* (Macquart 1844) is native to Australia but has spread worldwide (Baumgartner 1993). It was first introduced to the Western Hemisphere in Costa Rica in 1978 (Baumgartner and Greenberg 1984). Since then, it has spread into North America and has been documented in: Arizona (Baumgartner 1986), Arkansas (Meek et al. 1998), California (Greenberg 1988), Colorado (DeJong and Chadwick 1997), Florida (Mertins 1991), Illinois (Shahid et al. 2000), Louisiana (Martin et al. 1996), Michigan (Shahid et al. 2000), Nebraska (Figarola and Skoda 1998), New Mexico (Shahid et al. 2000), Oklahoma (Ahadizadeh et al. 2014), Ontario (Rosati and VanLaerhoven 2007), Tennessee (Shahid et al. 2000), Texas (Richard and Aherns 1993), West Virginia (Joy and D'Avanzo 2007) and Wisconsin (Marche 2013). Theoretical distributions of *C. rufifacies* include Indiana (Whitworth 2006, Rosati and VanLaerhoven 2007, Jones et al. 2019), but were not actually documented. Only one other *Chrysomya* species is known in Indiana: Picard (2013) documented one female adult *Chrysomya megacephala* (Fabricius 1794), on chicken liver. This was the first record of a species of *Chrysomya* in the state, being collected in Indianapolis, Indiana, approximately 240 km south of Valparaiso, Indiana. The authors corresponded with Purdue Entomological Research Collection (PERC) personnel to ensure that there are no *C. rufifacies* voucher specimens from the state, and there are none. This paper presents the first documented *C. rufifacies* in Indiana.

The field site where the collections occurred is located on Valparaiso University's campus in Valparaiso, Indiana (approximate coordinates, 41° 27′ 37.9″ N, 87° 03′ 03.3″ W). It has been used for decomposition studies using pigs as a carrion model, consistently in the summer and fall months since 2012. It is located in the clearing of a shady, wooded area in a remote part of campus. Research is conducted inside a metal dog kennel measuring 6.1 m × 36.1 m × 31.8 m to prevent predation.

Third instar larvae of *C. rufifacies* were collected on 26 September 2017. During the time of year, it was unseasonably warm, with temperatures reaching up to 37 °C and with average temperatures of 26.55 °C. Average humidity ranged from 60–78%, and there was no precipitation during the period of corpse exposure. Other larval species of Calliphoridae collected in the same trial include *Lucilia coeruleiviridis* Macquart (1855), *Lucilia sericata* (Meigen 1826), *Phormia regina* (Meigen 1826), *Calliphora vicina* Robineau-Desvoidy (1830), and *Calliphora vomitoria* (Linneaus 1758).

Identifications were made in the laboratory by Kristi Bugajski using Sukontason et al. (2008). The most distinguishing feature of *C. rufifacies* is a "hairy" appearance from distinct, elongated tubercles (Sukontason et al. 2008) (Fig. 1–2). This feature will help differentiate between *C. rufifacies* and *C. megacephala*. In addition to the tubercles, *C. rufifacies* has an incomplete and heavily pigmented peritreme with the middle slit being slightly bent (Sukontason et al. 2008). Adults of the two congeners can be separated using Whitworth's (2006) key.

The species was collected for a second time on 5 October 2019. Third instar larvae

**Figure 1**. Three third instar *Chrysomya rufifacies* collected from Valparaiso, Indiana on 26 September 2017.

were found on one of six pigs that were placed in the field on 1 October 2019. At the start of the trial, temperatures were above average, reaching 31.5 °C on the day of placement, which is consistent with when the species was previously found. Despite higher temperatures of up to 34 °C in September 2018, no specimens were found. They were also not found in 2019 prior to October.

The timing of the first collections of *Chrysomya* species, as reported in the literature, looks consistent in the Midwestern states and Canada. *Chrysomya rufifacies* were collected in Wisconsin on 2–3 October 2012 by Marche (2013). Rosati and VanLaerhoven (2007) found *C. rufifacies* in Windsor, Ontario on 7 October 2004. Likewise, Picard (2013) found the congener *C. megacephala* in Indiana on 24 September 2012.

One of the concerns about the non-native *C. rufifacies* is that it will outcompete native calliphorids, in particular *Cochliomyia macellaria* (Fabricius 1775) (Baumgartner 1993). However, if the current trend of only seeing *C. rufifacies* in the fall months continues, this will not have a large impact on the species composition in northwestern Indiana because *C. macellaria* is a warm-weather species that is not typically found in the fall. In eight years of research, *C. macellaria* has

**Figure 2**. Close up of the posterior end of a third instar *Chrysomya rufifacies* collected from Valparaiso, Indiana on 26 September 2017.

never been found in the fall months at this field site.

*Chrysomya* are warm-weather species, and it is probable that these distribution records are from migrating populations that are established in the southern states (Rosati and VanLaerhoven 2007). Rosati and VanLaerhoven (2007) provided maps with a projected range for *C. rufifacies* based on climate change trends that included most of the United States. This paper confirms their predictions that the species would be found in Indiana. This paper documents the first confirmation of *C. rufifacies* in the state, specifically Northwest Indiana. The authors suspect that it is present in other parts of the state if the conditions are adequate.

## Acknowledgments

The authors wish to thank the biology department at Valparaiso University, Birky Family Farms, Dr. Jeffrey Holland, and the PERC and the members of Team Maggot who helped with the research (Shelby Leucata, Monique LeDonne, and Lauren Smith).

## Literature Cited

**Ahadizadeh, E.N., H.R. Ketchum, and P.R. Mathews. 2014**. New geographic record of the hairy maggot blow fly, *Chrysomya rufifacies*, in Oklahoma. Southwestern Entomologist 39 (1): 191–192.

**Baumgartner, D.L. 1986.** The hairy maggot blow fly, *Chrysomya rufifacies* confirmed in Arizona. Journal of Entomological Science 21: 130–132.

**Baumgartner, D.L. 1993**. Review of *Chrysomya rufifacies* (Diptera, Calliphoridae). Journal of Medical Entomology 30 (2): 338–352.

**Baumgartner D.L., and B. Greenberg. 1984.** The genus *Chrysomya* (Diptera: Calliphoridae) in the New World. Journal of Medical Entomology 21: 105–113.

**DeJong, G.D., and J.W. Chadwick. 1997.** Additional county records and a correction to the checklist of the Calliphoridae (Diptera) of Colorado, with a new state record for *Chrysomya rufifacies*. Journal of the Kansas Entomological Society 70 (1): 47–51.

**Fabricius, J.C. 1775**. Systema entomologiae, sistens insectorum classes, ordines, genera, species, adiectis synonymis, locis, descriptionibus, observationibus. Officina Libraria, Kortii, Flensburgi et Lipsiae. 832 pp.

**Fabricius, J.C. 1794.** Entomologia systematica emendata et aucta. Tomus quartus. C. G. Proft, Fil. Et Soc., Hafniae [= Copenhagen]. 472 pp.

**Figarola, J.L.M., and S.R. Skoda. 1998**. *Chrysomya rufifacies* (Macquart) (Diptera: Calliphoridae) in Nebraska. Journal of Entomological Science 33: 319–321.

**Greenberg, B. 1988.** *Chrysomya megacephala* (F.) (Diptera: Calliphoridae) collected in North America and notes on *Chrysomya* species present in the New World. Journal of Medical Entomology 25: 199–200.

**Jones, N., T. Whitworth, and S.A. Marshall. 2019.** Blow flies of North America: Keys to the subfamilies and genera of Calliphoridae, and to the species of the subfamilies Calliphorinae, Luciliinae, and Chrysomyinae. Canadian Journal of Arthropod Identification 1–191.

**Joy, J.E., and V.A. D'Avanzo. 2007.** *Chrysomya rufifacies* **(**Diptera: Calliphoridae) from West Virginia. Proceedings of the West Virginia Academy of Science 63–66.

**Linnaeus, C. 1758.** Systema naturae per regna tria naturae. Tomus I. Editio decima, reformata. Laurentii Salvii, Holmiae. 824 pp.

**Macquart, J. 1844.** Diptères exotiques nouveaux ou peu connus. . [Tome deuxième, 3.e partie.]. Mémoires de la Société des Sciences, de l'Agriculture et des Arts, de Lille. 1842: 162–460.

**Macquart, J. 1855.** Diptères exotiques nouveaux ou peu connus. 5.e supplément. Mémoires de la Société royale des Sciences, de l'Agriculture et des Arts, de Lille, 2. Série 1: 25–156.

**Marche, J.D. 2013**. First Record of *Chrysomya rufifacies* (Diptera: Calliphoridae) in Wisconsin. Great Lakes Entomologist 46 (1-2): 135–137.

**Martin, C.S., C.E. Carlton, and C.L. Meek. 1996.** New distribution record of the hairy maggot blow fly *Chrysomya rufifacies*. Southwestern Entomologist 21: 477–478.

**Meek, C.L., C. Puskarich-May, and C.E. Carlton. 1998**. New State Record for the hairy maggot blow fly *Chrysomya rufifacies* (Macquart). Southwestern Entomologist 23 (4): 373–375.

**Meigen, J.W. 1826.** Systematische Beschreibung der bekannten europäischen zweiflügeligen Insekten. Fünfter Thiel. Schulzische Buchhandlung, Hamm. 412 pp.

**Mertins, J. 1991.** Blow fly infestation detected in new geographic areas, pp. 1–2. In Dx Monitor (Animal Health Report), American Association Veterinary Laboratory Diagnosticians, U.S. Animal Health Assoc. and USDA-APHIS, Winter issue.

**Picard, C.J. 2013**. First Record of *Chrysomya megacephala* Fabricius. (Diptera: Calliphoridae) in Indiana, USA. Proceedings of the Entomological Society of Washington 115 (3): 265–267.

**Richard, R.D., and E.H. Ahrens. 1993**. New distribution record for the recently introduced blow fly *Chrysomya rufifacies* (Macquart) in North America. Southwestern Entomologist 8: 216–218.

**Robineau-Desvoidy, J.B. 1830.** Essai sur les Myodaires. Mémoires. présentés par divers savans a l'Académie Royale des Sciences de l'Institut de France. Tome deuxième. Gauthier-Villars, Paris. 813 pp.

**Rosati, J.Y., and S.L. VanLaerhoven. 2007**. New Record of *Chrysomya rufifacies* (Diptera : Calliphoridae) in Canada: Predicted range expansion and potential effects on native species. Canadian Entomologist 139 (5): 670–677.

**Shahid, S.A., R.D. Hall, N.H. Haskell, and R.W. Merritt. 2000**. *Chrysomya rufifacies* (Macquart) (Diptera: Calliphoridae) established in the vicinity of Knoxville, Tennessee, USA. Journal of Forensic Science 45 (4): 896–897.

**Sukontason, K., S. Piangiai, S. Siriwattanarungsee, and K.L. Sukontason. 2008.** Morphology and development rate of blow flies *Chrysomya megacephala* and *Chrysomya rufifacies* in Thailand: Applications in forensic entomology. Parasitology Research 102 (6): 1207–1216.

**Whitworth, T. 2006.** Keys to the genera and species of blow flies of America north of Mexico. Proceedings of the Entomological Society of Washington 108 (3): 689–725.

# *Ailanthus altissima* Aqueous Extract Deters *Spodoptera frugiperda* Oviposition

Ryan L. Wagner* and Jordan A. Card

Department of Biology, Millersville University of Pennsylvania, 288 Roddy Hall, Millersville, PA 17551-0302

* Corresponding author: (e-mail: Ryan.Wagner@millersville.edu)

**Abstract**

*Ailanthus altissima* (Mill.) Swingle (Simaroubaceae), the tree of heaven, is a highly invasive tree species containing phytochemicals with a range of biological activities. Exposure to novel chemistry from invasive plant species may result in changes to established Lepidopteran behaviors, including feeding or oviposition. However, the impact of *A. altissima* chemistry on insect behavior has not been extensively explored. Therefore, *A. altissima* extract was tested for oviposition deterrence effects against *Spodoptera frugiperda* (J. E. Smith) (Noctuidae). Oviposition substrates were treated with water-soluble extractions of *Zea mays* (L), *A. altissima*, or a combination of both. Moths were then placed in chambers containing different substrate treatments and allowed to oviposit. Given choice, *S. frugiperda* demonstrated a higher percentage of egg deposition on *Z. mays* treated substrate compared to water treated substrate or *A. altissima* treated substrate. Significantly, when *Z. mays* treated substrate was subsequently treated with *A. altissima* extract, *S. frugiperda* was deterred from laying eggs on its preferred substrate (*Z. mays*) in a concentration-dependent manner. This observed change in behavior suggests that the deterrent properties of *A. altissima* phytochemicals may have economically important crop protection applications in controlling pest species like *S. frugiperda.*

**Keywords:** Oviposition, phytochemicals, behavioral deterrent, *Ailanthus*

---

Phytochemicals embedded in the leaf cuticle or within leaf tissues are known to play significant roles in plant host selection by Lepidoptera (Thompson and Pellmyr 1991, Renwick and Chew 1994). Host selection can be based on the presence of unique phytochemistry (Haribal et al. 1996), the concentration of select phytochemicals (Pereyra and Bowers 1988), or the relative proportions of different compounds within the tissue (Nishida et al. 1987). Host specific cues can positively influence behavior as seen by Meagher et al. (2011) where *Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae) preferentially selected grass when given a choice between corn, forage grass, and an artificial surface. Ultimately, the process of host selection for oviposition is relatively complex, potentially integrating multiple signals, to result in a behavioral choice (Lund et al. 2019).

With the establishment of invasive plant species into local ecosystems, opportunities exist to explore the impact of novel phytochemicals from these plants (Cappuccino and Arnason 2006) on oviposition. *Ailanthus altissima* (Mill.) Swingle (Simaroubaceae), the tree of heaven, is a highly successful invasive tree species (Mastelić and Jerković 2002) containing phytochemicals with a range of biological activities (Alves et al. 2014). Methylene chloride extracts from *A. altissima* leaves inhibited germination and growth of alfalfa, *Medicago sativa* L. (Fabaceae), and showed weak insecticidal properties against *Aedes aegypti* L. (Culicidae) (Tsao et al. 2002), while Pavela et al. (2014) demonstrated antifeedant activity in *Spodoptera littoralis* (Boisduval) associated with *A. altissima* methanol extracts. The potential use of novel phytochemicals from invasive plants, like *A. altissima,* to deter oviposition prior to host selection represents an exciting application for economically valuable crops.

One of the most widespread and damaging insect pests to cash crops is the fall armyworm, *S. frugiperda*, which feeds primarily on plants in the grass family (Nagoshi et al. 2012) with its most severe impact on corn, *Zea mays* L. (Poaceae) (Cruz et al. 1999). To reduce the damage caused by the fall armyworm and other crop pests, the United States spends over $10 billion annually on synthetic pesticides that, when applied, cause an increase in acute poisoning; cancer, and chronic diseases in humans; contaminated food products; destruction of

beneficial predators; pesticide resistance; honey bee poisoning; and crop damage (Pimentel 2005). Therefore, the development of alternative management resources is of great interest. The purpose of this research was to evaluate the biological properties of *A. altissima* with the specific objective of determining if its phytochemicals can influence Lepidopteran oviposition behavior.

*Spodoptera frugiperda* larvae were purchased from Benzon Research (Carlisle, PA) and reared at 21 °C with 16:8 h L:D cycles until pupation. Pupae were transferred by hand to individual 100 x 15 mm petri dishes until emergence. Newly emerged moths were kept isolated for two days to optimize fecundity (Rogers and Marti 1994).

*Ailanthus altissima* leaflets were collected from the campus of Millersville University and stored at –20°C. Four-week-old *Z. mays* was grown from seed in the Millersville University greenhouse (25°C, 14:10 h L:D cycles) and harvested immediately prior to extraction. Tissue (25 g) was pulverized with mortar and pestle then extracted with deionized, distilled water (100 ml) for one hour. The supernatant was clarified by centrifugation (5 min, 2,400 rpm, 21°C) and applied to oviposition substrates.

Twenty-six oviposition chambers were constructed using 950 ml mason jars containing cotton balls soaked in 10% sucrose as the moth food source. Cotton balls were replaced every 48 hours. Strips of chromatography paper (Whatman #1), 12 × 4 cm long, were saturated with their respective treatment, air dried for 2 h, and suspended into the chambers. Undiluted *A. altissima* extract contained 13.6 µg/µl residue with a final application concentration (1.0×) of 10.2 mg/strip. Control strips were treated with deionized water only. Each oviposition chamber represented one replicate.

Five male and five female *S. frugiperda* were introduced into each of thirteen oviposition chambers containing four different oviposition substrate treatments including: deionized water (control), *Z. mays* only, *A. altissima* only, and *Z. mays*/*A. altissima* combination. The *Z. mays*/*A. altissima* combination was prepared by sequentially treating the substrate in *Z. mays* extract, followed by *A. altissima* extract, with drying periods (2h) after each application. Thirteen chambers were prepared (as described) to test a concentration series whereby *A. altissima* extract concentrations of 0.0×, 0.25×, 0.5×, and 1.0× were applied onto substrates previously treated with *Z. mays* extract. The 1.0x concentration consisted of undiluted extract.

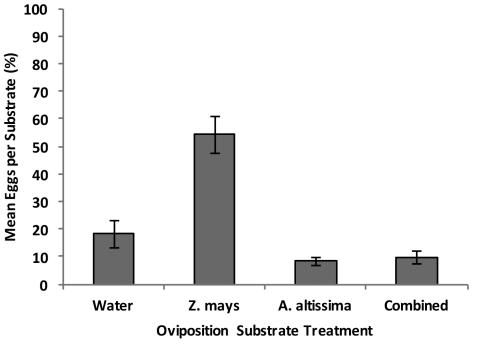


**Figure 1.** Mean (±SE) percentage of eggs laid by *S. frugiperda* after five days on oviposition substrates in a choice experiment. Oviposition substrates were prepared with four treatments including, water only, *Z. mays* extract only, *A. altissima* extract only, or *Z. mays* + *A. altissima* extract (combined) ($F = 18.33$; df = 3, 48; $P < .0001$).

After 5 days, substrates were collected and the eggs on each strip were counted and recorded. Eggs masses were examined by gently teasing apart egg mass layers to reveal individual eggs using an Olympus SZ3060 stereo microscope. Percent data were arcsin transformed and normality determined using an Anderson-Darling test. Differences in egg deposition were analyzed using one-way ANOVA and Tukey HSD post hoc test. Statistical analyses were completed using Microsoft Excel ® for Mac 2011 version 14.6.6.

A total of 9,220 eggs were found on oviposition strips across 13 oviposition chambers examining host preference (Fig. 1). *Spodoptera frugiperda* demonstrated significantly more egg deposition on substrate coated with *Z. mays* extract ($\bar{x} = 54.2\%$) compared to water ($\bar{x} = 18.3\%$) or *A. altissima* extract ($\bar{x} = 8.3\%$) ($F = 18.33$; df = 3, 48; $P < .0001$) (Fig. 1). *Zea mays* extract (alone) was preferred 6.5x more than the *A. altissima* extract (alone) and 5.5x more than the combined *Z. mays* + *A. altissima* extracts ($\bar{x} = 9.7\%$) (Fig. 1). A post hoc Tukey test showed no significant difference in oviposition activity exists when comparing the negative control (water) with *Z. mays* + *A. altissima* extract or *A. altissima* extract alone (Fig. 1).

When the two extracts were combined, egg deposition on *Z. mays* substrates decreased as the concentration of *A. altissima* extract increased ($F = 31.69$; df = 3, 48; $P < 0.001$) (Fig. 2). A post hoc Tukey test showed differences exist between 0.0x ($\bar{x} = 46.9\%$) and 0.25x ($\bar{x} = 25.6\%$) as well as 0.25x and 0.5x ($\bar{x} = 12.3\%$); no significant difference was present between the 0.5x and 1.0x ($\bar{x} =$

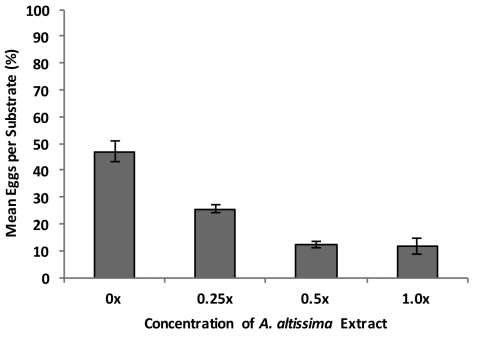


**Figure 2.** Mean (±SE) percentage of eggs laid by *S. frugiperda* on oviposition substrates treated with varying concentrations (0x, 0.25x, 0.5x, 1.0x) of *A. altissima* extract ($F = 31.69$; df = 3, 48; $P < .0001$). All oviposition substrates were saturated in *Z. mays* substrate and dried prior to application of *A. altissima* extract.

11.6%) concentrations. A total of 7,522 eggs were found on oviposition strips across the 13 oviposition chambers examining dose response (Fig. 2).

In this study, we demonstrated that the aqueous fraction of *A. altissima* contains phytochemicals capable of deterring *S. frugiperda* oviposition in a concentration-dependent manner, when applied to the preferred host substrate (Fig. 2). While it is possible that sequential treatment of the substrate diluted host phytochemical concentration, this is not anticipated as the twice saturated 0.0x control remained attractive to *S. frugiperda*. The decreased frequency of oviposition on *Z. mays* substrate in response to increasing concentrations of *A. altissima* extract suggests that the novel phytochemicals can directly deter oviposition, as seen in Senrung et al. (2014).

As a nocturnal ovipositor, *S. frugiperda* would not rely on visual cues; instead, volatile, tactile, or chemical contact cues would be more influential on oviposition behavior with tactile cues having been demonstrated to be highly important (Rojas et al. 2003). Interestingly, Rojas et al. (2003) reported that extracts of corn exhibited oviposition deterrent properties, while here we observed a preference for oviposition on corn-treated substrate. This difference in behavior may be due to the presence of different chemical combinations present in the water-soluble fraction here compared to the methanol and hexane fractions used by Rojas et al. (Pandey and Tripathi 2014).

Little is currently known regarding how manipulation of chemical metabolite profiles at sites of oviposition influences *S. frugiperda* behavior. While allelopathic activity by *A. altissima* has been extensively investigated (Heisey 1996, Heisey and Heisey 2003, Albouchi et al. 2013), the impact of its phytochemicals on oviposition behavior is less well understood. Through this work, evidence has been provided of previously unknown biological activity present in phytochemicals from *A. altissima* that detrimentally affects *S. frugiperda* oviposition behavior. Extracts from *A. altissima* have been demonstrated to reduce survivorship in pea aphid, *Acyrthosiphon pisum* (Harris) (De Feo et al. 2009) and reduce feeding activity/growth in *S. littoralis* (Pavela et al. 2014). Both De Feo et al. (2009) and Pavela et al. (2014) identified the quassinoid ailanthone as the biologically relevant phytochemical in *A. altissima*, however, neither group addressed oviposition behavior. While ailanthone is potentially present in the extract evaluated in this study, it was not chemically confirmed and it is not clear if this phytochemical could influence oviposition; thus further experiments on this metabolite are warranted.

Our demonstration of reduced oviposition is fundamental to the potential use of *A. altissima* as an alternative oviposition deterrent in an agricultural setting. Overuse of established pesticides has already led to resistance in Noctuidae agricultural pests, including *Spodoptera exigua* (Hübner) (Ahmad et al. 2018) and *Spodoptera litura* (F.) (Shad et al. 2012), emphasizing the need for new crop protection resources. Precedent clearly exists for phytochemical application (Isman 2006) as demonstrated by cypermethrin and permethrin, which both act as effective insecticidal and deterrent agents for *S. frugiperda* (Usmani and Knowles 2001). For example, crude hexane extracts (1% conc.) from curry leaf, *Murraya koenigii* (L.), reduced oviposition in the Noctuid *S. litura* by ~60% (Senrung et al. 2014). Further, essential oils present in cinnamon, clove, ginger, mint, and thyme significantly reduced oviposition (>80%) by the velvetbean caterpillar, *Anticarsia gemmatalis* (Hübner) when applied to host plants (Ribeiro et al. 2015). To our knowledge, this study represents the first evidence of *S. frugiperda* oviposition deterrence facilitated by phytochemicals from *A. altissima*. Future efforts to identify/isolate the semiochemical(s) present in *A. altissima* will potentially facilitate application in agricultural settings as a replacement for, or in conjunction with standard pesticides.

## Acknowledgments

This project was supported with funding from institutional sources including the Neimeyer-Hodgson Research Grant and the

Millersville University Faculty Academic Development Grants Program. The authors thank John R. Wallace for his guidance and critical feedback regarding this text.

## Literature Cited

**Ahmad, M., A. Farid, and M. Saeed. 2018.** Resistance to new insecticides and their synergism in *Spodoptera exigua* (Lepidoptera: Noctuidae) from Pakistan. Crop Protection 107:79–86.

**Albouchi, F., I. Hassen, H. Casabianca, and K. Hosni. 2013.** Phyochemicals, antioxidant, antimicrobial and phytotoxic activities of *Ailanthus altissima* (Mill.) Swingle leaves. South African Journal of Botany 87: 164–174.

**Alves, I.A.B.S., H. M. Miranda, L. A. L. Soares, and K. P. Randau. 2014.** Simaroubaceae family: botany, chemical composition and biological activities. Revista Brasileira de Farmacogosia 24: 481–501.

**Cappuccino, N., and J. T. Arnason. 2006.** Novel chemistry of invasive exotic plants. Biology Letters 2: 189–193.

**Cruz, I., M. L. C. Figueiredo, A. C. Oliveira, and C. A. Vasconcelos. 1999.** Damage of *Spodoptera frugiperda* (Smith) in different maize genotypes cultivated in soil under three levels of aluminum saturation. International Journal of Pest Management 45: 293–296.

**De Feo, V., E. Mancini, E. Voto, M. Curini, and C. Digilio. 2009.** Bioassay-oriented isolation of an insecticide from *Ailanthus altissima*. Journal of Plant Interactions 4: 119–123.

**Haribal, M., J. Alan, and A. Renwick, A. 1996.** Oviposition stimulants for the monarch butterfly: Flavonol glycosides from *Asclepias curassavica*. Phytochemistry 41: 139–144.

**Heisey, R. M. 1996.** Identification of an allelopathic compound from *Ailanthus altissima* (Simaroubaceae) and characterization of its herbicidal activity. American Journal of Botany 83: 192–200.

**Heisey, R. M., and T. K. Heisey. 2003.** Herbicidal effects under field conditions of *Ailanthus altissima* bark extract, which contains ailanthone. Plant and Soil 256: 85–99.

**Isman, M. B. 2006.** Botanical insecticides, deterrents, and repellents in modern agriculture and an increasingly regulated world. Annual Review of Entomology 51: 45–66.

**Lund, M., D. C., Brainard, and Z. Szendrei. 2019.** Cue hierarchy for host plant selection in *Pieris rapae*. Entomologia Experimentalis et Applicata 167: 330–340.

**Mastelić, J., and I. Jerković. 2002.** Volatile constituents from the leaves of young and old *Ailanthus altissima* (Mill.) Swingle tree. Croatica Chemica Acta 75: 189–197.

**Meagher, R. L., R. N. Nagoshi, and C. J. Stuhl. 2011.** Oviposition choice of two fall armyworm (Lepidoptera: Noctuidae) host strains. Journal of Insect Behavior 24: 337–347.

**Nagoshi, R.N., R. L. Meagher, M. Hay-Roe. 2012.** Inferring the annual migration patterns of fall armyworm (Lepidoptera: Noctuidae) in the United States from mitochondrial haplotypes. Ecology and Evolution 2: 1458–1467.

**Nishida. R., T. Ohsugi, S. Kokubo, and H. Fukami. 1987.** Oviposition stimulants of a *Citrus*-feeding swallowtail butterfly, *Papilio xuthus* L. Experientia 43: 342–344.

**Pandey, A., and S. Tripathi. 2014.** Concept of standardization, extraction and pre phytochemical screening strategies for herbal drug. Journal of Pharmacognosy and Phytochemistry 2: 115–119.

**Pavela, R., M. Zabka, T. Tylova, and Z. Kresinova. 2014.** Insecticidal activity of compounds from *Ailanthus altissima* against *Spodoptera littoralis* larvae. Pakistan Journal of Agricultural Science 51: 101–112.

**Pereyra, P. C., and M. D. Bowers. 1988.** Iridoid glycosides as oviposition stimulants for the buckeye butterfly, *Junonia coenia* (Nymphalidae). Journal of Chemical Ecology 14: 917–928.

**Pimentel, D. 2005.** Environmental and economic costs of the application of pesticides primarily in the United States. Environment, Development and Sustainability 7: 229–252.

**Renwick, J. A. A., and F. S. Chew. 1994.** Oviposition behavior in Lepidoptera. Annual Review of Entomology 39: 377–400.

**Riberio, R. C., T. V. Zanuncio, F. de S. Ramalho, C. A. D. da Silva, J. E. Serrão, and J. C. Zanuncio. 2015.** Feeding and oviposition of *Anticarsia gemmatalis* (Lepidoptera: Noctuidae) with sublethal concentrations of ten condiments essential oils. Industrial Crops and Products 74: 139–143.

**Rogers, C. E., and O. G. Marti Jr. 1994.** Reproductive potential of once-mated moths of the fall armyworm (Lepidoptera: Noctuidae). The Florida Entomologist 77: 402–410.

**Rojas, J. C., A. Virgen, and L. Crus-López. 2003.** Chemical and tactile cues influencing oviposition of a generalist moth, *Spodoptera frugiperda* (Lepidoptera: Noctuidae). Environmental Entomology 32: 1386–1392.

**Senrung, A., J. Singh, S. Sharma, T. N. Bhutia, and A. K. Singh. 2014.** Effect of *Murraya koenigii* extracts on feeding and ovipositional response of *Spodoptera litura* (Fab.) (Lepidoptera: Noctuidae). Journal of Entomological and Zoology Studies 2: 27–31.

**Shad, S. A., A. H. Sayyed, S. Fazal, M. A. Saleem, S. M. Zaka, and M. Ali. 2012.** Field evolved resistance to carbamates, organophosphates, pyrethroids, and new chemistry insecticides in *Spodoptera litura* Fab. (Lepidoptera: Noctuidae). Journal of Pest Science 85: 153–162.

**Thompson, J. N., and O. Pellmyr. 1991.** Evolution of oviposition behavior and host preference in Lepidoptera. Annual Review of Entomology 36: 65–89.

**Tsao, R., F. E. Romanchuk, C. J. Perterson, and J. R. Coats. 2002.** Plant growth regulatory effect and insecticidal activity of the extracts of the tree of heaven (*Ailanthus altissima* L.). BMC Ecology 2(1). **doi:** 10.1186/1472-6785-2-1

**Usmani, K.A., and C. O. Knowles. 2001.** Toxicity of pyrethroids and effect of synergists to larval and adult *Helicoverpa zea*, *Spodoptera frugiperda*, and *Agrotis ipsilon* (Lepidoptera: Noctuidae). Journal of Economic Entomology 94: 868–873.

# Buprestidae, Cerambycidae, and Siricidae Collected in Baited Funnel Traps on Drummond Island, Chippewa County, Michigan

Robert A. Haack

USDA Forest Service, Northern Research Station, 3101 Technology Blvd., Suite F, Lansing, MI 48910 (e-mail: robert.haack@usda.gov (emeritus))

**Abstract**

Trapping of bark- and wood-infesting insects in 2015 and 2016 at multiple locations on Drummond Island in northern Lake Huron, using baited multi-funnel traps, yielded 4 species of Buprestidae, 24 Cerambycidae, and 4 Siricidae. In 2015, all funnel traps were baited with the plant volatiles α-pinene, ethanol, and cis-3-hexenol, and were either black or green in color, and placed either at heights similar to the lower canopy of nearby trees (4-5 m) or at 1-2 m above groundline. In 2016, all traps were green, hung at 1-2 m above groundline, and baited with the cerambycid pheromones fuscumol acetate, monochamol, and syn-(2,3)-hexanediol. In 2015, 29 species of Buprestidae, Cerambycidae, and Siricidae were captured, compared with 9 cerambycids in 2016. The original target insect, emerald ash borer, *Agrilus planipennis* Fairmaire (Coleoptera: Buprestidae), an invasive pest from Asia, was not trapped in either year.

**Keywords:** Borers, *Agrilus planipennis*, funnel trap, lures, trap height, trap color

---

Drummond Island is Michigan's second largest island, covering about 130 square miles (≈ 83,131 ac or 33,642 ha) with 58% being state-owned and managed by the Michigan Department of Natural Resources as part of the Lake Superior State Forest (Karen Rodock, Michigan DNR, personal communication). Drummond Island lies near the northwestern end of Lake Huron and is the easternmost point of Michigan's Upper Peninsula (Fig. 1a). The bedrock on Drummond Island is largely limestone and dolomite (a magnesium rich limestone), and in many areas is exposed at the surface, forming broad, flat expanses known as alvars that are covered with little or no soil and usually dominated by grasses and sedges (Lincoln 2018).

Several tree species occur on Drummond Island and other nearby islands. The most common conifers include balsam fir [*Abies balsamea* (L.) Mill.], tamarack [*Larix laricina* (Du Roi) K. Koch], white spruce [*Picea glauca* (Moench) Voss], black spruce [*Picea mariana* (Mill.) Britton, Sterns & Poggenburg], red pine (*Pinus resinosa* Sol. ex Aiton), white pine (*Pinus strobus* L.), northern white cedar (*Thuja occidentalis* L.), and eastern hemlock [*Tsuga canadensis* (L.) Carrière]. Similarly, the most common hardwood tree species include red maple (*Acer rubrum* L.), sugar maple (*A. saccharum* Marshall), yellow birch (*Betula alleghaniensis* Britton), paper birch (*B. papyrifera* Marshall), American beech (*Fagus grandifolia* Ehrh.), white ash (*Fraxinus americana* L.), black ash (*F. nigra* Marshall). green ash (*F. pennsylvanica* Marshall), American hophornbeam [*Ostrya virginiana* (Mill.) K.Koch], balsam poplar (*Populus balsamifera* L.), bigtooth aspen (*P. grandidentata* Michaux), trembling aspen (*P. tremuloides* Michx.), northern red oak (*Quercus rubra* L.), and American basswood (*Tilia americana* L.) (Scharf and Chamberlin 1978, Weatherbee 2014, MDNR 2015, Reznicek et al. 2016, Lincoln 2018).

In 2015, at the time this study began, the Asian buprestid known as the emerald ash borer (EAB, *Agrilus planipennis* Fairmaire) had not yet been found on Drummond Island, but populations were nearby in both parts of Michigan and Ontario, Canada. EAB was first detected in North America in 2002 near Detroit, Michigan and Windsor, Ontario (Haack et al. 2002, 2015), and has now spread to 35 US states and 5 Canadian provinces as of June 2020 (EAB Info 2020). EAB was first detected in Michigan's Upper Peninsula in 2005 on the shores of Lake Superior in Brimley State Park, Chippewa County (Storer et al. 2009; Fig. 1a). In 2008, EAB was detected in Sault Ste. Marie, Ontario, located near the northern end of the St. Marys River, which connects Lake Superior to Lake Huron (Fig. 1a). In 2009, EAB was detected across the St. Marys River in Sault Ste. Marie, MI. In nearby areas, EAB was next reported on Sugar Island (Michigan) in

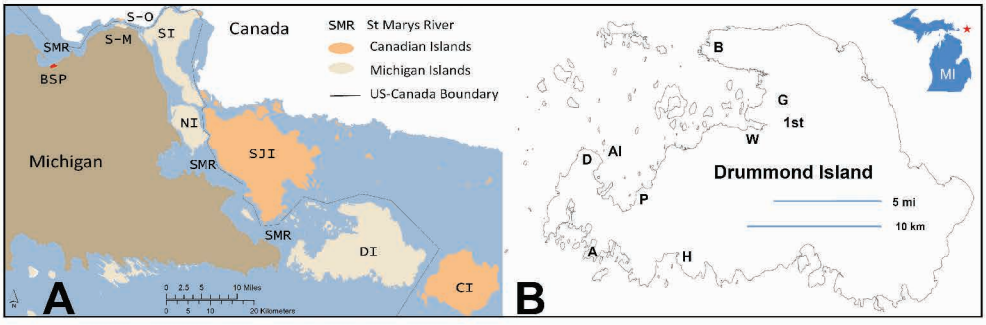


**Figure 1. A**. Outline map of Drummond Island (DI), surrounding islands and mainland of Michigan and Ontario, Canada. See text for historical spread of emerald ash borer in this area. Codes are: BSP = Brimley State Park; CI = Cockburn Island; NI = Neebish Island; SI = Sugar Island; SJI = St Joseph Island; SMR = St. Marys River; S-M = Sault Ste. Marie, MI; and S-O = Sault Ste. Marie, Ontario. **B**. Outline map of Drummond Island and surrounding islands that comprise Drummond Island Township, Michigan, indicating the 2015 and 2016 trapping locations, which were from north to south, Bruce Point (B), GEMS (Grouse Enhanced Management Systems; G), boat launch site to First Lake (1st), Wa-Wen Resort (W), Arrow Island (AI), Dix Point (D), Township Park (P), Anderson Point (A), and Helen Lake (H). Drummond Island is to the left of the red asterisk in the inset map of Michigan.

2012 and then on St Joseph Island (Ontario) in 2014 (Fig. 1a).

The initial objective of the present study in 2015 was to monitor for presence of EAB on Drummond Island by setting out baited funnel traps at various locations around the island. In 2016 another survey took place, but smaller in scale, using funnel traps with different baits. In this paper a list is presented of the bark- and wood-boring Buprestidae, Cerambycidae, and Siricidae that were collected during the 2-year study.

**Methods: 2015 Study.** Six trapping sites were selected in 2015, five on Drummond Island itself and one on Arrow Island (AI; N 46.0236° Lat, W 83.8239° Long), a small (2.5 ac, 1 ha), state-owned island near (0.4 km) the west side of Drummond Island (Fig. 1b). The other sites were generally located on the eastern side of Drummond Island, anticipating that if EAB were to fly over water to Drummond Island they would come from infestations to the west in Michigan's Upper Peninsula or from the north on St. Joseph Island (Fig. 1a). The northernmost trapping site was located on Bruce Point (B; N 46.0908°, W 83.7194°) on state land with traps placed in trees near the shoreline (Fig. 1b). Moving southward, traps were placed along the forest edge at Wa-Wen Resort (W; N 46.0332°, W 83.6756°) near the shoreline of Potagannissing Bay. More inland, traps were placed along the forest edge of the Potagannissing River on state land near the parking lot where boats are launched to enter First Lake (1st; N 46.0416°, W 83.6544°). The next trapping site was along the forest edge at Dix Point (D; N 46.0251°, W 83.8419°) on state land. The last trapping site was located on the southwestern side of Drummond Island along the shoreline forest edge of Anderson Point (A; N 45.9576°, W 83.8431°) near Fairview Cove, where the author has a cabin. Ash trees were common at four of the trapping sites (AI, B, 1st, and W) but rare at the other two sites (A and D; Fig. 1b).

Traps were deployed during 4–6 June 2015 and taken down on 3 October 2015 (≈ 120 trapping days). The traps were checked irregularly, usually every 3–4 weeks. A kayak was used to reach the traps on Arrow Island. At each site, four 12-unit funnel traps (Contech Enterprises Inc., Victoria, British Columbia, Canada) were deployed. Two of the traps had black funnels and two had green funnels like those pictured in Petrice and Haack (2015). The funnels of all traps were coated with Fluon (Northern Products Inc., Woonsocket, RI), a slippery substance that improves trapping efficiency (Graham et al. 2010). Two traps, one of each color, were suspended from branches in the lower canopy of dominant trees so that the bottom of the collection cup was about 4-5 m above groundline. The other two traps, one of each color, were suspended in the understory, using lower branches of trees so that the collection cup was about 1 m above groundline. Typically, the two traps of the same color were suspended from the same tree or two trees within 5 m of each other. The two other traps were suspended from one or two trees that were at least 10 m away from the first pair. Traps were suspended from ash trees at four sites (1st, AI, B, and W), but not the other two sites (A and

D) where no ash trees were nearby. Most of the selected ash trees had some branch dieback, but none had signs of EAB infestation (e.g., EAB adult exit holes or EAB larval galleries; Haack et al. 2015). In all cases, traps were suspended so that they were clearly visible, and any interfering branches were pruned. The base of the collection cups had a small screen funnel to allow rainwater to drain. A circular piece of window screening was fitted snugly near the bottom of each cup to suspend captured insects above any moisture that accumulated. Pieces of No-Pest Strip (Spectrum Group, St. Louis, MO) were placed inside each collection cup to quickly kill trapped insects. Dichlorvos, an organophosphate insecticide, is the active ingredient in No-Pest Strips.

Three different attractant lures were attached to the inside of each funnel trap and changed every 6–7 weeks. The lures included one α-pinene UHR (ultra-high release) pouch with a release rate of 2.3 g per day at 26 °C (Alpha Scents, Inc., West Linn, OR), one ethanol UHR pouch with a release rate of about 300 mg per day at 20 °C (Contech Enterprises Inc., Delta, BC), and two cis-3-hexenol bubble caps with a combined release rate of 7.4 mg per day at 20 °C (Contech Enterprises Inc). The α-pinene and ethanol lures are common attractants for a wide range of bark- and wood-infesting insects (Millar and Hanks 2017, Rabaglia et al. 2019). The volatile leaf alcohol *cis*-3-hexenol serves as a host attractant for EAB adults (Poland et al. 2019). In addition, at Anderson Point (A), a single unbaited black funnel trap was deployed in the lower canopy near the other traps as a blank control.

**2016 Study.** Six trapping sites were used in 2016, including three of the original sites (A, D, and W; Fig. 1b). The three new sites, from north to south, were along the forest edge at the GEMS (Grouse Enhanced Management Systems) site on state land (G; N 46.0580°, W 83.6622°), a forested area of the Drummond Island Township Park that had recently been selectively logged (P; N 45.9884°, W 83.7850°), and in a forest opening near Helen Lake on state land (H; N 45.9547°, W 83.7514°; Fig. 1b). A single Fluon-coated, green, 12-unit funnel trap was deployed at each site. The 2016 study was not focused on EAB and therefore sites with a different tree mixture were used: *Abies* and *Populus* dominated the GEMS site, *Acer* and *Fagus* the Township Park, and *Pinus* the Helen Lake site.

Traps were deployed on 23 May 2016 and taken down on 20 October 2016 (148 trapping days). The traps were checked irregularly, every 3–6 weeks. Traps were suspended from lower tree branches so that the collection cup was about 1 m above groundline. The collection cups were prepared as in 2015. The lures used on each trap included one fuscumol acetate bubble cap with a release rate of 2 mg/day at 20 °C (Synergy Semiochemicals Corporation, Delta, BC), one monochamol bubble cap with a release rate of ~750 ug/day at 20 °C (Synergy), and one syn-(2,3)-hexanediol (a racemic mix of six-carbon diols) bubble cap with an undetermined release rate (Synergy). These compounds are known pheromones or attractants for several cerambycid species in multiple cerambycid subfamilies that infest both conifers and hardwoods (Millar and Hanks 2017), and they can be suspended from the same traps with no apparent antagonistic effects on attraction to cerambycids (Fan et al. 2019). In addition, at the Anderson Point (A) site, a single unbaited green funnel trap was deployed in the lower canopy near the other trap as a blank control.

**Insect sorting and identification.** In both years, all insects in the collection cups were emptied into labeled plastic zip-lock bags and kept frozen until sorted. Insects were later removed from one bag at a time and placed in a Petri dish and viewed under a dissecting microscope. All specimens of Buprestidae, Cerambycidae, and Siricidae were removed and placed in labelled vials or pinned. These insects were identified using various reference books and keys, including Wellso et al. (1976) and Paiero et al. (2012) for Buprestidae; Yanega (1996) and Lingafelter (2007) for Cerambycidae, and Schiff et al. (2006, 2012) for Siricidae. Identifications were later confirmed by comparing specimens with previously identified insects in the Michigan State University (MSU), Department of Entomology, Albert J. Cook Arthropod Research Collection in East Lansing, MI. Currently, the borers identified in this paper reside in the author's personal collection but later will be placed in the MSU collection.

**Results: 2015 Study.** Overall, 129 borers (4.5 borers per 100 trap-days) were collected in the 24 baited traps in 2015, including 7 buprestids (4 species, but no EAB), 115 cerambycids (21 species), and 7 siricids (4 species) (Table 1). No borers were collected in the single unbaited control trap. Of the 21 cerambycid species, five were members of the subfamily Cerambycinae, six were Lamiinae, eight were Lepturinae, and two were Spondylidinae (Table 1). Nine of the 21 cerambycid species develop primarily in conifers (softwoods), 11 mostly in broadleafed trees (hardwoods), and the larval hosts of one cerambycid species are still unknown: *Idiopidonia pedalis* (LeConte) (Table 1). Similarly, one of the buprestid species de-

**Table 1. Details on the bark- and wood-infesting Buprestidae, Cerambycidae, and Siricidae collected in baited funnel traps on Drummond Island in 2015 and 2016.**

| Family, Subfamily, Species | Year(s) when collected | Number of adults collected: 2015 B / G[1] | 2015 U / C[2] | Total for 2015 | Total for 2016 | Sites where collected in 2015–16[3] | Primary months collected in 2015–16[4] | Common larval hosts[5] |
|---|---|---|---|---|---|---|---|---|
| CERAMBYCIDAE | | 67 / 48 | 58 / 57 | 115 | 53 | | | |
| Cerambycinae | | | | | | | | |
| *Clytus ruricola* (Olivier) | 2015 | 0 / 3 | 2 / 1 | 3 | 0 | 1st W | VII-VIII | Hardwoods |
| *Cyrtophorus verrucosus* (Olivier) | 2015 | 1 / 1 | 0 / 2 | 2 | 0 | B W | VI-VII | Hardwoods |
| *Microclytus gazellula* (Haldeman) | 2015 | 1 / 2 | 1 / 2 | 3 | 0 | D W | VI | Hardwoods |
| *Neoclytus acuminatus* (Fab.) | 2016 | - | - | 0 | 1 | W | VI-VII | Hardwoods |
| *Pronocera collaris* (Kirby) | 2015 | 1 / 0 | 0 / 1 | 1 | 0 | W | VI-VII | Conifers |
| *Xylotrechus undulatus* (Say) | 2015 | 12 / 12 | 9 / 15 | 24 | 0 | 1st A AI D W | VI-VIII | Conifers |
| Lamiinae | | | | | | | | |
| *Acanthocinus pusillus* Kirby | 2015-16 | 2 / 2 | 2 / 2 | 4 | 1 | 1st D P | VI-VIII | Conifers |
| *Aegomorphus modestus* (Gyllenhal) | 2015-16 | 1 / 0 | 0 / 1 | 1 | 1 | H W | VI-VIII | Hardwoods |
| *Astylopsis collaris* (Haldeman) | 2015-16 | 2 / 2 | 2 / 2 | 6 | 2 | D G W | VI-VIII | Hardwoods |
| *Graphisurus fasciatus* (DeGeer) | 2016 | - | - | 0 | 6 | P | VII-VIII | Hardwoods |
| *Monochamus marmorator* Kirby | 2015-16 | 0 / 1 | 0 / 1 | 2 | 1 | A G | VII-VIII | Conifers |
| *Monochamus notatus* (Drury) | 2015-16 | 2 / 0 | 2 / 0 | 3 | 1 | A H | VIII-IX | Conifers |
| *Monochamus scutellatus* (Say) | 2015-16 | 10 / 10 | 10 / 10 | 59 | 39 | 1st A G H P W | VI-IX | Conifers |
| Lepturinae | | | | | | | | |
| *Bellamira scalaris* (Say) | 2015 | 2 / 0 | 0 / 2 | 2 | 0 | 1st W | VII-VIII | Hardwoods |
| *Evodinus monticola* (Randall) | 2016 | - | - | 0 | 1 | A | V-VI | Conifers |
| *Grammoptera subargentata* (Kirby) | 2015 | 1 / 0 | 1 / 0 | 1 | 0 | B | IX | Hardwoods |
| *Hyperplatys aspersa* (Say) | 2015 | 1 / 0 | 1 / 0 | 1 | 0 | AI | VII-VIII | Hardwoods |
| *Idiopidonia pedalis* (LeConte) | 2015 | 1 / 0 | 1 / 0 | 1 | 0 | W | VI-VII | Unknown |
| *Pidonia ruficollis* (Say) | 2015 | 0 / 2 | 1 / 1 | 2 | 0 | W | VI | Hardwoods |
| *Rhagium inquisitor* (L.) | 2015 | 2 / 1 | 2 / 1 | 3 | 0 | 1st A W | VI-VIII | Conifers |
| *Trachysida mutabilis* (Newman) | 2015 | 1 / 3 | 4 / 0 | 4 | 0 | 1st D W | VI-VIII | Hardwoods |
| *Trigonarthris minnesotana* (Casey) | 2015 | 20 / 0 | 12 / 8 | 20 | 0 | 1st AI B W | VI-VIII | Hardwoods |

**Table 1. Continued.**

| Family, Subfamily, Species | Year(s) when collected | Number of adults collected: 2015 B / G[1] | 2015 U / C[2] | Total for 2015 | Total for 2016 | Sites where collected in 2015–16[3] | Primary months collected in 2015–16[4] | Common larval hosts[5] |
|---|---|---|---|---|---|---|---|---|
| Spondylidinae | | | | | | | | |
| *Asemum striatum* (L.) | 2015 | 6 / 8 | 7 / 7 | 14 | 0 | D, W | VI-VII | Conifers |
| *Tetropium cinnamopterum* Kirby | 2015 | 1 / 1 | 1 / 1 | 2 | 0 | D W | VI-VII | Conifers |
| BUPRESTIDAE | | 4 / 3 | 3 / 4 | 7 | 0 | | | |
| *Agrilus anxius* Gory | 2015 | 0 / 1 | 1 / 0 | 1 | 0 | W | VII-VIII | *Betula* |
| *Buprestis maculativentris* Say | 2015 | 1 / 0 | 1 / 0 | 1 | 0 | 1st | VIII | Conifers |
| *Dicerca tenebrica* (Kirby) | 2015 | 2 / 1 | 0 / 3 | 3 | 0 | W | VI-VII | *Populus* |
| *Poecilonota cyanipes* (Say) | 2015 | 1 / 1 | 1 / 1 | 2 | 0 | W | VIII-IX | *Populus* |
| SIRICIDAE | | 7 / 0 | 4 / 3 | 7 | 0 | | | |
| *Sirex nigricornis* Fabricius | 2015 | 1 / 0 | 1 / 0 | 1 | 0 | A | IX | Conifers |
| *Urocerus albicornis* (Fabricius) | 2015 | 3 / 0 | 2 / 1 | 3 | 0 | 1st D | VII-VIII | Conifers |
| *Urocerus cressoni* Norton | 2015 | 1 / 0 | 0 / 1 | 1 | 0 | 1st | VIII | Conifers |
| *Xeris melancholicus* (Westwood) | 2015 | 2 / 0 | 1 / 1 | 2 | 0 | B | VII-IX | Conifers |

[1]Trap color: B = black, G = green.
[2]Trap location: U = understory, C = canopy.
[3]Trapping site codes given in Fig. 1b.
[4]Months: V = May, VI = June, VII = July, VIII = August, IX = September.
[5]Common hosts are based on Gosling 1973, 1983, 1984, 1986; Gosling and Gosling 1977; Heffern et al. 2015; Lingafelter 2007; Paiero et al. 2012; Schiff et al. 2006, 2012; Wellso et al. 1976; Yanega 1996.

veloped in conifers and three in hardwoods (Table 1). All four siricid species developed in conifers (Table 1).

Ten of the 29 borer species were each represented by only a single individual, and six others by only two individuals each (Table 1). By contrast, only four of the 29 borer species were collected in numbers of 10 individuals or greater, all of which were cerambycids, including *Asemum striatum* (L.) (14 individuals), *Monochamus scutellatus* (Say) (20), *Trigonarthris minnesotana* (Casey) (20), and *Xylotrechus undulatus* (Say) (24; Table 1). Of the six trapping sites, Arrow Island had the fewest with only 3 borer species collected (all cerambycids), whereas Wa-Wen Resort had the most, with 19 species collected (3 buprestids and 16 cerambycids).

There were no striking differences between the diversity of borers collected in traps placed in the understory (catching 22 of the 29 species) compared with those placed in the lower canopy (catching 21 of the 29 species). For the four cerambycid species that were captured most frequently, they were captured at equal or nearly equal numbers in traps placed at the two height positions: 7 of 14 *A. striatum* were captured at the lower position ($\chi 2$ test, $P = 1$), and similarly, 10 of 20 *M. scutellatus* ($P = 1$), 11 of 20 *T. minnesotana* ($P = 0.655$), and 13 of 24 *X. undulatus* ($P = 0.683$).

Considering trap color, the buprestids and cerambycids were captured in both black and green traps, whereas all siricids were captured in black traps. Overall, three of the four buprestid species and 18 of the 21 cerambycid species were captured in black traps, compared with three buprestid and 13 cerambycid species being collected in green traps. Of the four most frequently collected cerambycids, only *T. minnesotana* showed a strong color preference with all 20 individuals collected in black traps ($\chi 2$ test, $P < 0.0001$). By contrast, the others were captured at equal or nearly equal numbers in black and green traps: *A. striatum* ($P = 0.593$), *M. scutellatus* ($P = 1$), and *X. undulatus* ($P = 1$).

Borers were captured during each of the five trapping periods. Overall, 25 individual borers (9 species) were collected during the first collecting period that ended on 23 June 2015, 41 (16 species) on 12 July, 45 (13 species) on 15 August, 14 (7 species) on 1 September, and 4 (4 species) on 3 October. Of the 29 borer species collected, the cerambycid *Microclytus gazellula* (Haldeman) was only captured during the first sampling period in June, whereas the cerambycid *Grammoptera subargentata* (Kirby) and siricid *Sirex nigricornis* Fabricius were only captured in the last sampling period that occurred mostly in September and ended in early October (Table 1).

**2016 Study.** Overall, 53 borers (6.0 borers per 100 trap-days), representing 9 cerambycid species, were collected in the six baited traps used in 2016 (Table 1). No buprestids or siricids were trapped in 2016, and no borers were collected in the single unbaited control trap. Moreover, of the nine cerambycid species, one was a member of the subfamily Cerambycinae, seven were Lamiinae, and one was a Lepturinae (Table 1). Six of the nine cerambycid species collected in 2016 had been captured in 2015 and three were new (Table 1). Four of the nine 2016 cerambycid species developed primarily in hardwoods and five in conifers (Table 1).

Six of the nine 2016 cerambycid species were represented by only a single individual, one was collected twice, another six times, and one (*M. scutellatus*) 39 times. No borers were collected at Dix Point (D) in 2016. At the other five 2016 sites, usually only two or three cerambycid species were captured throughout the entire trapping season, and only *M. scutellatus* was collected at all five sites (Table 1).

From 6 to 19 borers were captured during each of the first five trapping periods, but none were collected during the last period (12 September to 20 October 2016). Overall, 6 individual borers (2 species) were collected during the first collecting period that ended on 4 June 2016, 10 (3 species) on 4 July, 7 (4 species) on 20 July, 19 (4 species) on 21 August, and 11 (2 species) on 12 September. Besides *M. scutellatus*, the only other cerambycid collected during the first collection period that ended on 4 June was *Evodinus monticola* (Randall), and similarly *Monochamus notatus* (Drury) was the only other borer trapped during 21 August to 12 September (Table 1).

**Discussion.** The original impetus for this study was to sample for EAB on Drummond Island. However, no EAB adults were collected in either 2015 or 2016. Moreover, as of June 2020, EAB has still not been reported on Drummond Island, nor on nearby Neebish Island (Michigan) to the northwest or Cockburn Island (Ontario) to the southeast (Fig. 1a). Nevertheless, EAB is becoming more common on the mainland of Michigan's eastern Upper Peninsula, as well as on the mainland of Ontario between Sault Ste. Marie and Manitoulin Island, which is directly east of Cockburn Island. EAB was first detected on Manitoulin Island in 2011. Given that EAB-infested areas generally surround Drummond Island, EAB will likely arrive someday, either by active flight or being assisted by humans, such as

by transporting infested firewood (Haack et al. 2010). In 2018, Michigan's internal EAB quarantine was repealed given that EAB was known to occur in all but four of Michigan's 83 counties at that time (MDARD 2018). As a result, movement of ash trees, ash logs, and ash firewood was no longer regulated within Michigan, although the public was still encouraged not to move firewood long distances. Therefore, inadvertent movement of EAB-infested ash trees, logs, or firewood could easily bring EAB to Drummond Island as well as other nearby islands.

All 32 species of borers collected in this study (4 buprestids, 24 cerambycids, and 4 siricids) were already reported to occur in Michigan (Gosling 1973, Wellso et al. 1976, Gosling and Gosling 1977, Schiff et al. 2012). Of these borers, county occurrence data in Michigan have only been published for the cerambycids (Gosling 1973, 1983, 1984; Gosling and Gosling 1977). In these four papers, county-level data are presented for 228 cerambycid species, of which 104 species were recorded in Michigan's Upper Peninsula. Using the above four papers as the baseline, seven of the 24 cerambycid species reported in this paper [*Aegomorphus modestus* (Gyllenhal), *Astylopsis collaris* (Haldeman), *Grammoptera subargentata* (Kirby), *Hyperplatys aspersa* (Say), *Microclytus gazellula* (Haldeman), *Monochamus marmorator* Kirby, and *Pidonia ruficollis* (Say)] are new county records for Chippewa County, MI, and two of these species are recorded for the first time in Michigan's Upper Peninsula (*A. collaris* and *M. gazellula*). Undoubtedly many more borer species occur on Drummond Island than were collected in this study, considering that Michigan has at least 120 species of buprestids (Wellso et al. 1976; Haack et al. 2002, 2009; Maier 2012; MacRae and Basham 2013), 228 species of cerambycids (Gosling 1973, 1983, 1984; Gosling and Gosling 1977), and 10 species of siricids (Schiff et al. 2012).

Although no striking differences in borer diversity were found between understory and canopy traps in the present 2015 study, other researchers have found significant differences relative to trapping height (Dodds 2014, Rassati et al. 2019, Ulyshen and Sheehan 2019). Similarly, there were few striking differences in buprestid and cerambycid attraction to traps of different colors in the present study (black vs. green), although others have reported that trap color did affect borer diversity and abundance. For example, many adult buprestids show a preference for green traps over black traps (Petrice and Haack 2015, Skvarla and Dowling 2017) as well as green over purple traps (Rassati et al. 2019). For cerambycids, Rassati et al. (2019) found that trap color (green vs. purple) significantly affected diversity and abundance at the cerambycid subfamily level and for certain individual cerambycid species. This was true in present study as well, with black traps preferred by the lepturine cerambycid *T. minnesotana*, and similarly by the cerambycine *Xylotrechus colonus* (Fabricius) (Skvarla and Dowling 2017). In other studies, black traps were shown to be superior to clear traps or white traps in catching various buprestids, cerambycids, and siricids (de Groot and Nott 2001, Hurley et al. 2015, Allison and Redak 2017). In a study in South Africa, where six traps of each color were used, more *Sirex noctilio* F. adults were captured in black intercept panel traps (33 individuals; Alpha Scents, West Linn, OR) and black 12-unit funnel traps (28), compared with either light blue (6) or dark blue (2) funnel traps (Brett P. Hurley, University of Pretoria, South Africa, unpublished data).

The lures used in the present study were dramatically different between years, with plant volatiles (α-pinene, ethanol, and cis-3-hexenol) used in 2015 and cerambycid pheromones (fuscumol acetate, monochamol, and syn-(2,3)-hexanediol) used in 2016. Many borers use plant volatiles to locate their host plants, especially those that infest trees stressed by environmental factors such as drought (Mattson and Haack 1987, Haack and Petrice 2019). Environmental stress often increases ethanol production in plant tissues in both hardwoods and conifers, and similarly the production of the resin monoterpene α-pinene in stressed conifers (Kimmerer and Kozlowski 1982, Mattson and Haack 1987, Turtola et al. 2003). Ethanol and α-pinene, especially when combined, are well known attractants to many bark- and wood-infesting insects, including many cerambycids and siricids (Miller 2006, Johnson et al. 2013, Millar and Hanks 2017, Hanks et al. 2018). The ethanol and α-pinene lures used in 2015 were likely responsible for attracting most of the wood borers collected that year. By contrast, the cis-3-hexenol lures were specifically used to attract EAB, given that this alcohol is released from hardwood trees, including ash foliage, and is known to be attractive to EAB adults (Poland et al. 2019). Nevertheless, no EAB were collected in 2015, suggesting that EAB had not yet invaded Drummond Island, or if present, was not near any of the six 2015 trapping sites.

Of the nine cerambycid species collected in 2016, when cerambycid pheromones were used as lures, two species are known to use fuscumol acetate as a pheromone component [*Aegomorphus modestus* and *Graphisurus fasciatus* (DeGeer)]; three use monochamol (*Monochamus marmorator*, *M.*

*notatus* and *M. scutellatus*); and one uses syn-(2,3)-hexanediol [*Neoclytus acuminatus* (Fab)], (Millar and Hanks 2017, Hanks et al. 2018, Meier et al. 2020). No pheromone information was found for the three other cerambycids collected in 2016 (*Acanthocinus pusillus* Kirby, *Astylopsis collaris*, and *Evodinus monticola*).

There are likely many additional borer species on Drummond Island than were collected in this study, given that relatively few lures and trapping techniques were employed as well as relatively few habitats were sampled. In addition to the genera of the tree species listed in the introduction of this paper, many other trees and large shrubs occur on Drummond Island and nearby islands, including native species of *Alnus, Amelanchier, Celastrus, Corylus, Cornus, Crataegus, Dirca, Ilex, Juniperus, Lonicera, Malus, Myrica, Prunus, Rhus, Rhamnus, Ribes, Rosa, Rubus, Salix, Sambucus, Shepherdia, Sorbus, Symphoricarpos, Taxus,* and *Ulmus* (Scharf and Chamberlin 1978, Weatherbee 2014, Reznicek et al. 2016, Lincoln 2018). Borers may also use some of the island's smaller shrubs and woody vines as larval hosts, such as species of *Arctostaphylos*, *Diervilla, Gaultheria*, *Hypericum*, *Rhododendron* (formerly *Ledum*), *Spiraea, Vaccinium* and *Vitis*. For example, the Cerambycinae *Cyrtophorus verrucosus* (Olivier), two of which were collected in this study (Table 1), are 7–11 mm long and have been reared from many species of hardwood trees (Yanega 1996, Lingafelter 2007), and also woody shrubs in the genera *Cornus, Shepherdia*, and *Vaccinium* (Heffern et al. 2018), all of which grow on Drummond Island.

## Acknowledgments

The author thanks Karen Rodock (Michigan DNR) for permission to survey on State land on Arrow Island and Drummond Island, and for data on state land acreage on Drummond Island; Erin Bullas-Appleton (Canadian Food Inspection Agency) for providing EAB survey information for Ontario; Helen Enander (Michigan Natural Features Inventory) for creating the base maps used in this publication; Lawrence Hanks (University of Illinois at Urbana-Champaign) for providing background information on cerambycid pheromones; Brett P. Hurley (University of Pretoria, South Africa) for providing unpublished data, and Davide Rassati (University of Padua, Italy), Toby R. Petrice (USDA Forest Service, Northern Research Station), and two anonymous reviewers for comments on an earlier draft of this paper. This research was supported in part by the USDA Forest Service.

## Literature Cited

**Allison, J. D., and R. A. Redak. 2017.** The impact of trap type and design features on survey and detection of bark and woodboring beetles and their associates: a review and meta-analysis. Annual Review of Entomology 62: 127–146.

**de Groot, P., and R. Nott. 2001.** Evaluation of traps of six different designs to capture pine sawyer beetles (Coleoptera: Cerambycidae). Agricultural and Forest Entomology 3: 107–111.

**Dodds, K. J. 2014.** Effects of trap height on captures of arboreal insects in pine stands of northeastern United States of America. Canadian Entomologist 146: 80–89.

**EAB Info. 2020.** Emerald ash borer information network. Online at: http://www.emeraldashborer.info/

**Fan, J. T., O. Denux, C. Courtin, A. Bernard, M. Javal, J. G. Millar, L. M. Hanks, and A. Roques. 2019.** Multi-component blends for trapping native and exotic longhorn beetles at potential points-of-entry and in forests. Journal of Pest Science 92: 281–297.

**Gosling, D. C. L. 1973.** An annotated list of the Cerambycidae of Michigan (Coleoptera) part I, introduction and the subfamilies Parandrinae, Prioninae, Spondylinae, Aseminae, and Cerambycinae. The Great Lakes Entomologist 6: 65–84.

**Gosling, D. C. L. 1983.** New state records of Cerambycidae from Michigan (Coleoptera). The Great Lakes Entomologist 16: 187.

**Gosling, D. C. L. 1984.** Cerambycid host plants in a southwestern Michigan woodland (Coleoptera: Cerambycidae). The Great Lakes Entomologist 17: 69–78.

**Gosling, D. C. L. 1986.** Ecology of the Cerambycidae (Coleoptera) of the Huron Mountains in Northern Michigan. The Great Lakes Entomologist 19: 153–162.

**Gosling, D. C. L., and N. M. Gosling. 1977.** An annotated list of the Cerambycidae of Michigan (Coleoptera). Part II, the subfamilies Lepturinae and Lamiinae. The Great Lakes Entomologist 10: 1–37.

**Graham, E. E., R. F. Mitchell, P. F. Reagel, J. D. Barbour, J. G. Millar, and L. M Hanks. 2010.** Treating panel traps with a fluoropolymer enhances their efficiency in capturing cerambycid beetles. Journal of Economic Entomology 103: 641–647.

**Haack, R. A., and T. R. Petrice. 2019.** Historical population increases and related inciting factors of *Agrilus anxius*, *Agrilus bilineatus*, and *Agrilus granulatus liragus* (Coleoptera: Buprestidae) in the Lake States (Michigan,

Minnesota, and Wisconsin). The Great Lakes Entomologist 52: 21–33.

**Haack, R. A., E. Jendek, H. Liu, K. R. Marchant, T. R. Petrice, and T. M. Poland. 2002.** The emerald ash borer: a new exotic pest in North America. Newsletter of Michigan Entomological Society 47: 1–5.

**Haack, R. A., T. R. Petrice, and J. E. Zablotny. 2009.** First report of the European oak borer, *Agrilus sulcicollis* (Coleoptera: Buprestidae), in the United States. The Great Lakes Entomologist 42: 1–7.

**Haack, R. A., T. R. Petrice, and A. C. Wiedenhoeft. 2010.** Incidence of bark- and wood-boring insects in firewood: a survey at Michigan's Mackinac Bridge. Journal of Economic Entomology 103: 1682–1692.

**Haack, R. A., Y. Baranchikov, L. S. Bauer, and T. M. Poland. 2015.** Emerald ash borer biology and invasion history, pp. 1-13. In R. G. Van Driesche and R. C. Reardon (eds.), Biology and control of emerald ash borer. U.S. Department of Agriculture, Forest Service, Forest Health Technology Enterprise Team, Morgantown, WV.

**Hanks, L. M., J. A. Mongold-Diers, T. H. Atkinson, M. K. Fierke, M. D. Ginzel, E. E. Graham, T. M. Poland, A. B. Richards, M. L. Richardson, and J. G. Millar. 2018.** Blends of pheromones, with and without host plant volatiles, can attract multiple species of cerambycid beetles simultaneously. Journal of Economic Entomology 111: 716–724.

**Heffern, D. J., J. Vlasak, and R. L. Alten. 2018.** Larval host plant records, distributional records, and biological information on North American Cerambycidae (Coleoptera). The Coleopterists Bulletin 72: 739–750.

**Hurley, B. P., J. Garnas, and M. F. Cooperband, 2015.** Assessing trap and lure effectiveness for the monitoring of Sirex noctilio. *Agricultural* and Forest *Entomology* 17: 64–70

**Johnson, C. W., J. R. Meeker, W. G. Ross, S. D. Petty, B. Bruce, and C. Steiner. 2013.** Detection and seasonal abundance of *Sirex nigricornis* and *Eriotremex formosanus* (Hymenoptera: Siricidae) using various lures and trap trees in central Louisiana, U.S. Journal of Entomological Science 48: 173–183.

**Kimmerer, T. W., and T. T. Kozlowski. 1982.** Ethylene, ethane, acetaldehyde, and ethanol production by plants under stress. Plant Physiology 69: 840–847.

**Lincoln, J. M. 2018.** Ecological review of Maxton Plains alvar. Michigan Natural Features Inventory Report Number 2018-24, Lansing, MI. 28 pp.

**Lingafelter, S. W. 2007.** Illustrated key to the longhorned wood-boring beetles of the eastern United States. Coleopterists Society, Special Publication 206, North Potomac, MD.

**MacRae, T. C., and J. P. Basham. 2013.** Distributional, biological, and nomenclatural notes on Buprestidae (Coleoptera) occurring in the U.S. and Canada. The Pan-Pacific Entomologist 89: 125–142.

**Maier, C. 2012.** New distributional records of invasive Coleoptera found in Connecticut and other Eastern states: *Agrilus cyanescens* (Ratezburg) (Buprestidae), *Lilioceris lilii* (Scopoli), and *Pyrrhalta viburni* (Paykull) (Chrysomelidae). Proceedings of the Entomological Society of Washington 114: 501–512.

**Mattson, W. J., and R. A. Haack. 1987.** The role of drought in outbreaks of plant–eating insects. BioScience 37: 110–118.

**Meier, L. R., Y. Zou, J. A. Mongold-Diers, J. G. Millar, and L. M. Hanks. 2020.** Pheromone composition and chemical ecology of six species of cerambycid beetles in the subfamily Lamiinae. Journal of Chemical Ecology 46: 30–39.

**MDARD (Michigan Department of Agriculture and Rural Development). 2018.** Online at: https://www.michigan.gov/documents/mdard/Michigan_EAB_Interior_Quarantine_Notice_of_Repeal_634689_7.pdf

**MDNR (Michigan Department of Natural Resources). 2015.** Drummond Island comprehensive resource management plan. Michigan Department of Natural Resources, Wildlife Division Report No. 3612. Online at: https://www.michigan.gov/documents/dnr/drummond_island_management_plan_498925_7.pdf

**Millar, J. G., and L. M. Hanks. 2017.** Chemical ecology of cerambycids, pp. 161–196. In Q. Wang (ed.), Cerambycidae of the world: biology and pest management. CRC Press/Taylor & Francis, Boca Raton, FL.

**Miller, D. R. 2006.** Ethanol and (–)-α-pinene: attractant kairomone for some large wood-boring beetles in southeastern USA. Journal Chemical Ecology 32: 779–794.

**Paiero, S. M., M. D. Jackson, A. Jewiss-Gaines, T. Kimoto, B. D Gill, and S. A Marshall. 2012.** Field guide to the jewel beetles of northeastern North America. Canadian Food Inspection Agency, Ottawa, Ontario, Canada

**Petrice, T. R., and R. A. Haack 2015.** Comparison of different trap colors and types for capturing adult *Agrilus* (Coleoptera: Buprestidae) and other Buprestids. The Great Lakes Entomologist 48: 45–66.

**Poland T. M., T. R. Petrice, and T. M. Ciaramitaro. 2019.** Trap designs, colors, and lures for emerald ash borer detection. Frontiers in Forests and Global Change 2: 80. Doi: 10.1126/science.1257570

**Rabaglia, R. J., A. I. Cognato, E. R. Hoebeke, C. W. Johnson, J. R. Labonte, M. E. Carter, and J. J. Vlache. 2019.** Early detection and rapid response: a ten-year summary of the USDA Forest Service program of surveillance for non-native bark and ambrosia beetles. American Entomologist 65: 29–42.

**Rassati, D., L. Marini, M. Marchioro, P. Rapuzzi, G. Magnani, R. Poloni, F. Di Giovanni, P. Mayo, and J. Sweeney. 2019.** Developing trapping protocols for wood-boring beetles associated with broadleaf trees. Journal of Pest Science 92: 267–279.

**Reznicek, A. A., E. G. Voss, and B. S. Walters. 2016.** Michigan flora online. Online at: https://michiganflora.net/

**Scharf, W. C., and M. L. Chamberlin. 1978.** Ecological inventory of Harbor Island Chippewa County, Michigan. Unpublished report submitted to The Nature Conservancy, Minneapolis, MN. 17 pp.

**Schiff, N. M., Valley, S. A., LaBonte, J. R. and Smith, D. R. 2006.** Guide to the siricid woodwasps of North America. USDA Forest Service, Forest Health Technology Enterprise Team, Morgantown, West Virginia, FHTET–2006–15. 102 pp.

**Schiff, N. M., H. Goulet, D. R. Smith, C. Boudreault, A. D. Wilson, and B. E. Scheffler. 2012.** Siricidae (Hymenoptera: Symphyta: Siricoidea) of the Western Hemisphere. Canadian Journal of Arthropod Identification 21: 1–305.

**Skvarla, M. J., and A. P. G. Dowling. 2017.** A comparison of trapping techniques (Coleoptera: Carabidae, Buprestidae, Cerambycidae, and Curculionoidae excluding Scolytinae). Journal of Insect Science 17(1): 1–28.

**Storer, A. J., J. M. Marshall, M. M. Philip, D. G. McCullough, and N. W. Siegert. 2009.** The history of emerald ash borer discoveries in the Upper Peninsula of Michigan from 2005 to 2007, p. 77. In K. A. McManus and K. W. Gottschalk (eds.), Proceedings, 19th U.S. Department of Agriculture Interagency Research Forum on Invasive Species 2008. 8-11 January 2008, Annapolis, MD. General Technical Report NRS-P-36. U.S. Department of Agriculture, Forest Service, Northern Research Station, Newtown Square, PA.

**Turtola, S., A. M. Manninen, R. Rikala, and P. Kainulainen. 2003.** Drought stress alters the concentration of wood terpenoids in Scots pine and Norway spruce seedlings. Journal of Chemical Ecology 29: 1981–1995.

**Ulyshen, M. D., and T. N. Sheehan. 2019.** Trap height considerations for detecting two economically important forest beetle guilds in southeastern US forests. Journal of Pest Science 92: 253–265.

**Weatherbee, E. E. 2014.** The flora of Cockburn Island, Ontario, Canada. The Michigan Botanist 53: 73–136.

**Wellso, S. G., G. V. Manley, and J. A. Jackman. 1976.** Key and notes on the Buprestidae (Coleoptera) of Michigan. The Great Lakes Entomologist 9: 1–22.

**Yanega, D. 1996.** Field guide to northeastern longhorned beetles (Coleoptera: Cerambycidae). Illinois Natural History Survey Manual 6, Champaign. IL.

# INSTRUCTIONS FOR AUTHORS

## SUBJECTS

Papers dealing with any aspect of entomology will be considered for publication in The Great Lakes Entomologist. Appropriate subjects are those of interest to professional and amateur entomologists in the North Central States and Canada, as well as general papers and revisions directed to a larger audience while retaining an interest to readers in our geographic area. All manuscripts are refereed by at least two reviewers.

## SUBMISSION REQUIREMENTS

Manuscripts must be typed in English with line numbers, double-spaced, with 1" margins and uploaded. Please use italics rather than underline. Use subheadings sparingly and set them into paragraphs in boldface.

Footnotes (except for authors' addresses, which must be on the title page, and treated as a footnote), legends, and captions of illustrations should be on separate pages.

Titles should be concise, identifying the order and family discussed. The author of insect species must be given fully at least once in the abstract and text, but not in the title. If a common name is used for a species or group, it should be in accordance with the common names published by the Entomological Society of America, or the Entomology Society of Canada.

The format for references must follow that described in the style guidelines used by the Entomological Society of America, except that journal titles are not abbreviated.

Please make sure that all references cited in the text appear in the Literature Cited section, and that all references in the Literature Cited appear in the body of the paper.

## FIGURES AND TABLES

Photographs should be high resolution digital files (300 dpi). Drawings, charts, graphs, and maps must be scaled to permit proper reduction without loss of detail.

Figures must meet the following criteria: (1) gray-scale images must be submitted with a resolution of at least 300 dpi ; (2) line art or graphs must be sent as 600 dpi.

Scanned images should be saved in the native application. Never embed the images in a word-processing document.

Captions for figures should be numbered consecutively and typed in order at the end of the manuscript. Captions should not be attached to illustrations.

Tables should be kept as uncluttered as possible, and should fit normally across a page when typeset.

Tables cannot be submitted as Excel files or graphics, but only as text. Contributors should follow the Council of Biology Editors Style Manual, 7th ed., and examine recent issues of The Great Lakes Entomologist for proper format of manuscripts.

The organizational format for a manuscript is as seen in the recent issues of the journal: TITLE, Author(s), Abstract, Introduction, Methods & Materials, Results, Discussion, Acknowledgments, Literature Cited, Tables, and a List of Figures.

Do not use extra spaces between paragraphs or references in the Lit. Cited.

The columns of text in tables should be aligned with TABS, not spaces.

Some symbols may not translate properly from one computer system to another. Do not use extbols.

## PAGE CHARGES

Papers published in The Great Lakes Entomologist are subject to a page charge of $20 per published page for non-members and $5 per published page for members. Color images can be included at no additional charge. Members of the Society, who are authors without funds from grants, institutions, or industry, and are unable to pay costs from personal funds, may apply to the Society for financial assistance. Application for subsidy must be made at the time a manuscript is initially submitted for publication. Authors will receive a page proof, together with a page charge form. Reprints will be provided as PDF files. Extensive changes to the proof by the author will be billed at a rate of $1.00 per line.

## COVER ARTWORK

Cover art or photographs are desired for upcoming issues. They are published free of charge. We only require that they be suitably prepared as described for images above, and that the subject be identified as accurately as possible.

## ATTRIBUTION AND USE POLICIES

Reproduction, posting, transmission or other distribution or use of the article or any material therein, in any medium as permitted by a personal-use exemption or by written agreement of The Great Lakes Entomologist, requires credit to The Great Lakes Entomologist as original publisher (e.g., The Great Lakes Entomologist © 2017).

Personal-use Exceptions: The following uses are always permitted to the author(s) and do not require further permission from ValpoScholar provided the author does not alter the format or content of the articles, including the copyright notification:

- Posting on ResearchGate, Academia.edu, or a similar website;
- Storage and back-up of the article on the author's computer(s) and digital media (e.g., diskettes, back-up servers, Zip disks, etc.), provided that the article stored on these computers and media is not readily accessible by persons other than the author(s);
- Posting of the article on the author(s) personal website, provided that the website is non-commercial;
- Posting of the article on the internet as part of a non-commercial open access institutional repository or other non-commercial open access publication site affiliated with the author(s)'s place of employment (e.g., a Entomology professor at the University of the Great Lakes can have her article appear in the University of the Great Lakes's Department of Entomology online publication series); and
- Posting of the article on a non-commercial course website for a course being taught by the author at the university or college employing the author.

People seeking an exception, or who have questions about use, should contact the editors.

## GENERAL TERMS AND CONDITIONS OF USE

Users of the ValpoScholar website and/or software agree not to misuse the ValpoScholar service or software in any way.

The failure of ValpoScholar to exercise or enforce any right or provision in the policies or the Submission Agreement does not constitute a waiver of such right or provision. If any term of the Submission Agreement or these policies is found to be invalid, the parties nevertheless agree that the court should endeavor to give effect to the parties' intentions as reflected in the provision, and the other provisions of the Submission Agreement and these policies remain in full force and effect. These policies and the Submission Agreement constitute the entire agreement between ValpoScholar and the Author(s) regarding submission of the Article.

## EDITOR'S ADDRESS

All manuscripts for The Great Lakes Entomologist should be submitted via the online system at http://scholar.valpo.edu/tgle/ . Any questions regarding submissions should be sent to the Lead Scientific Editor, Kristi Bugajski, Valparaiso University, 1610 Campus Drive East Valparaiso, IN 46383. (email: kristi.bugajski@valpo.edu).

OTHER BUSINESS Other correspondence should be directed to the Secretary, Michigan Entomological Society, 2104 Needham Rd., Ann Arbor, MI 48104.

The Great Lakes Entomologist
Published by the Michigan Entomological Society

Volume 53 Nos. 1 & 2

ISSN 0090-0222

Table of Contents



Cover photo
*Karshomyia caulicola* (Coquillett)
Photo by
Joseph Moisan-De Serres